JOURNAL OF AGRICULTURAL RESEARCH

VOL. 61 WASHINGTON, D. C., SEPTEMBER 15, 1940 No. 6

EFFECT OF EXCHANGE SODIUM ON THE MOISTURE EQUIVALENT AND THE WILTING COEFFICIENT OF SOILS[1]

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INTRODUCTION

Laboratory research by numerous investigators has shown that sodium adsorbed by the clay complex of soils increases dispersion, pH values, swelling, osmotic imbibition, migration velocity, apparent density of puddled soils, and the hardness of dry aggregates; and lowers heat of wetting, sticky point, and permeability.

The moisture equivalent (*3, 4*)[5] and wilting coefficient (*5*), more than any other soil constants, have come into extensive use as reference points for the water relations of soils and plants. Sharp and Waynick (*17*), Joseph (*10*), Anderson (*1*), and others (*20*) have been in agreement in finding that adsorbed sodium markedly increases the moisture equivalent. Veihmeyer and Hendrickson (*21*) have concluded, however, that the change, when any, can be attributed to soil puddling. In connection with all results it can be appropriately mentioned that the magnitude of the effect that adsorbed sodium has on soil characteristics is related to the amount and character of the clay of the soil, to the extent to which other ions are replaced by sodium, and to the extent to which flocculating electrolytes are removed.

No one has reported on the effects of adsorbed sodium on the availability of moisture to plants in the wilting range, and, so far as the writers have been able to learn, no investigations have been conducted.

In agreement with laboratory findings, it has been extensively observed that when lands are irrigated with water containing a high proportion of sodium relative to the concentrations of other bases the permeability tends to be reduced. Accompanying the effects of adsorbed sodium on permeability are other adverse consequences, such as the accumulation in the root zone of the salt constituents of irrigation waters, increased erodibility, the loss of good tilth, and consequent unsatisfactory seedbeds. It is a common observation that

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[4] The authors gratefully acknowledge their indebtedness to George Y. Blair, assistant pomologist, Division of Irrigation Agriculture, for the major portion of the measurements of the wilting points reported in this paper.

[5] Italic numbers in parentheses refer to Literature Cited, p. 424.

soils with much adsorbed sodium are less permeable to rain water than they are to saline irrigation waters.

The expression "percent sodium," as used by Eaton (7) to designate the relation

$$\text{Sodium-total base ratio} = \frac{\text{milliequivalents Na} \times 100}{\text{milliequivalents of total bases}},$$

has come to be regarded as an important water-quality characterization. Because of the diversity of factors that bear on permeability and exchange reactions and the quantitative relationships involved, it has not been possible to closely delimit the concentrations and proportions of sodium in natural waters that should be avoided or designated as undesirable.

The relationships between adsorbed sodium and moisture equivalent that are reported on in this paper were investigated for the purpose of exploring the possibility of utilizing the moisture equivalent as an index to sodium-induced dispersion. As a part of the inquiry, comparisons were made between calcium-treated and sodium-treated soils with respect to (1) the effect of the acceleration rate of the centrifuge on the moisture equivalent; (2) the migration and segregation of sand, silt, and clay in dispersed soils during centrifuging; (3) the distribution of moisture in Ca soils and Na soils after centrifuging; (4) the effect of flocculation by electrolytes on the moisture equivalent of Na soils; and (5) the quantity of sodium that significantly affected moisture equivalent values when Ca and Na soils were mixed in different proportions. Finally, moisture equivalent and hydroscopicity comparisons were made between soils saturated with calcium, magnesium, sodium, and potassium. This was done for the purpose of determining whether the effects of potassium most closely resembled those of calcium and magnesium or those of sodium.

The relationships between the adsorbed sodium and moisture availability were investigated with 12 soils, both at the wilting coefficient and at the ultimate wilting point. The resulting data, together with the moisture equivalent values, are graphically presented with the use of a pF scale.[6] (See fig. 3.)

DESCRIPTION OF SOILS, DEFINITIONS, AND METHODS

The source, classification, and certain physical constants of the 12 soils used for these investigations are reported in table 1.

MOISTURE EQUIVALENT

The moisture-equivalent values represent the percentage of moisture remaining in 30-gm. samples of soil that had been saturated for 24 hours, drained for 30 minutes, and then centrifuged for 30 minutes in standard cups in a standard moisture equivalent centrifuge drum operated at a rotational speed of 2,440 r. p. m.[7] The centrifuge came to full speed in 15 seconds. Any free water remaining on the surface of the soil after centrifuging was removed by inverting the cups for several minutes and then wiping their inside walls with blotting paper.

[6] The pF value of soils is the logarithm of the equivalent capillary tension expressed in centimeters of water column.

[7] The centrifuge drum was belt-driven from a 1-hp. polyphase induction motor following a principle of operation worked out by E. S. Babcock, of Riverside, Calif. By the use of a large motor with a low connected load, the speed of the drum is determined alone by the cycles of the alternating current and the ratio of the two pulleys. The centrifuge drum was mounted on the vertical pulley, within which were two roller bearings set on a 1¼-inch shaft anchored in a concrete pedestal. With this machine the acceleration rate is rapid, though it is possible to reduce it by installing an autotransformer (choke coil) in the line.

TABLE 1.—*Series, source, and certain physical constants of 12 California soils used in the investigation reported herein*

Soil No.	Soil type	Location	Moisture equivalent [1]	Xylene equivalent [2]	Wilting coefficient [3]	Apparent density [1]	Exchange capacity per 100 gm.	Mechanical analysis of composite [1 4]		
								Sand, 2–0.05 mm.	Silt, 0.05–.002 mm.	Clay, 0.002 mm.
						Gm./Cc.	*Milli-equivalents*	*Percent*	*Percent*	*Percent*
279	Gold Ridge fine sandy loam	Sonoma County, sec. 35, T. 6 N., R. 9 W	10.6	4.4	3.6	1.31	3.34	71.3	22.2	6.5
1	Sierra loam	Riverside County, sec. 22, T. 2 S., R. 5 W	8.8	4.3	3.7	1.44	4.82	60.7	33.4	5.9
4	Yolo loam	Ventura County, sec. 26, T. 4 N., R. 20 W	11.5	4.3	4.7	1.43	7.13	64.6	26.2	9.2
2	Hanford sandy loam	Kern County, sec. 8, T. 31 S., R. 30 E	11.7	4.2	5.5	1.36	8.54	72.6	17.8	9.6
6	Yolo loam	Ventura County, sec. 26, T. 3 N., R. 22 W	12.4	5.3	5.7	1.46	8.02	62.2	28.7	9.1
3	do	Ventura County, sec. 2, T. 3 N., R. 21 W	14.1	4.0	5.8	1.41	9.32	50.7	37.3	12.0
9	Antioch clay loam	Stanislaus County, sec. 31, T. 3 S., R. 7 E	18.0	7.9	10.2	1.37	14.05	38.0	36.3	25.7
5	Yolo silt loam	Ventura County, sec. 19, T. 3 N., R. 21 W	20.6	7.5	9.8	1.27	16.46	40.5	41.1	18.4
10	Antioch clay	Stanislaus County, sec. 31, T. 3 S., R. 7 E	21.0	11.1	12.0	1.40	15.83	19.4	50.5	30.1
8	Yolo fine sandy loam	Ventura County, sec. 4, T. 2 N., R. 22 W	22.4	11.6	11.7	1.22	14.82	27.5	51.5	21.0
7	Rincon loam, silty phase	Ventura County, sec. 23, T. 3 N., R. 22 W	22.9	6.0	11.4	1.38	20.00	37.1	35.6	27.3
278	Aiken clay loam	Butte County, sec. 14, T. 22 N., R. 3 E	36.2	24.7	22.0	1.00	14.77	37.6	35.8	26.6

[1] Untreated soil.
[2] Means of determinations on washed, calcium-treated, and sodium-treated soils (percentage by weight).
[3] Ca soil.
[4] The mechanical analyses were made in accordance with the pipette method of the former Bureau of Chemistry and Soils, U. S. Department of Agriculture.

XYLENE EQUIVALENT

The xylene technique was the same as that used for the moisture equivalent, except that the samples were oven-dried just before they were wet with xylene. Xylene-equivalent measurements were made on washed, calcium-treated, and sodium-treated soils. The character of the adsorbed cation, in accordance with Joseph's (*10*) findings for soil clays, was without effect on the percentage of xylene held against a force of 1,000 times gravity. The greatest single departure between the xylene equivalents of any soil as treated with calcium and sodium was 2.6. The average of the values of the xylene equivalents was 12.9 for the 12 Ca soils and also for the 12 Na soils.

WILTING COEFFICIENT AND ULTIMATE WILTING POINT

The exhaustion of soil water that accompanies the wilting of plants, as recognized by Briggs and Shantz (*5*) and subsequently by others, is progressive, and extends through what is now termed the "wilting range." The fact that wilting does occur over a range of moisture percentages for a given soil adds to the difficulties associated with the selection of a uniform end point. The wilting coefficient, as here used, represents a degree of wilting from which only about one-third of the leaves of sunflowers recovered when the plants were placed in a dark, humid chamber overnight. Except as noted in table 10, each wilting coefficient and ultimate wilting point value is the average of six determinations.

Water and soil were alternately added (washed, Ca, and Na soils alike) as the cans were filled for the wilting coefficient measurements. After germination of the sunflower seeds additional water was added to the soil surface until a plant of the desired size was developed, after which the openings in the covers of the cans were closed with cotton. After the plants had been wilted and cropped, the soils were dried at 105° to 110° C. for 4 days. The soil of each can was then broken up and passed through a 2-mm. screen and rolled in a paper to remix. One sample was withdrawn from each wilting coefficient can for moisture equivalent measurement. Any moisture equivalent determination that appeared out of line was repeated with a new sample. The soil of the six cans of each treatment was then composited as stock samples for use in other parts of the investigation.

The "ultimate wilting point," a term suggested by Taylor, Blaney, and McLaughlin (*18*), is used in the present work to designate the moisture content at which the rate of movement of water into a 15- to 30-cm. sunflower plant, after wilting, from a 600-gm. mass of soil was only sufficient to bring about the recovery of the terminal pair of leaves (1 to 2 cm. long) when the plant was placed overnight in the moist chamber. As this end point was approached the loss of water was a fraction of a gram per day, or in the order of 0.1 percent of the weight of the soil mass. The atmospheric conditions in the Riverside, Calif., greenhouse where the plants were grown were conducive to high transpiration rates. After recording the pot weights at the wilting coefficient, the same plants were reexposed in the greenhouse until the ultimate wilting point was reached.

The old leaves of plants on the majority of the Na soils deteriorated somewhat more rapidly than those on the Ca soils, and the death of leaves on the former plants was sometimes accompanied by marginal

burning. Equally good distributions of roots were observed in the Ca and Na soils. A further reference is made to the wilting measurements under soil treatments.

APPARENT DENSITY

Apparent density of untreated soils, as reported in table 1, represents the weight per unit volume of air-dry soil that had been passed through a 2-mm. sieve. The volume was measured in a 5 by 5 by 2.5 cm. box. The box was filled and lightly tapped on the desk top and leveled off before weighing; the method of filling the boxes was as nearly alike as possible for all soils.

EXCHANGE CAPACITY

For the measurements of the exchange capacity, a 20-gm. portion of soil was digested with 250 cc. of neutral normal ammonium acetate and leached free of calcium with 500 cc. of neutral normal ammonium acetate. The excess ammonium acetate was then removed by leaching with neutral normal ammonium chloride followed by leaching with neutral methyl alcohol until the leachate was free from chloride ion. The adsorbed ammonia (NH_3) was distilled, in the presence of magnesia (MgO), into a 2-percent solution of boric acid and titrated with standard sulfuric acid. The values reported in table 1 are the means of washed, sodium-treated, and calcium-treated soils; the values were in close agreement.

PREPARATION OF SOILS

Two sets of soils were prepared for these investigations. The first set—designated washed, Ca, and Na soils—provided material for the comparisons of the effects of calcium and sodium on the moisture equivalent and on the wilting coefficient and ultimate wilting point. The second set—designated Ca, Mg, K, and Na soils—was used only for the supplementary hydroscopicity and moisture equivalent measurements where the effect of potassium was of principal interest. Descriptions of the methods employed in preparation of the two sets of soils follow.

WASHED, CALCIUM-TREATED, AND SODIUM-TREATED SOILS, AND MEASUREMENT OF EXCHANGE SODIUM

Three 4-kg. portions of each of the 12 soils (table 1) were weighed into porcelain dishpans. The washed soils were successively treated with distilled water in parallel with the soils treated with calcium and sodium. The Ca and Na soils were treated three successive times with 3-liter portions of normal calcium chloride and sodium chloride solutions, respectively. Each suspension was stirred several times during the day and allowed to settle overnight; the supernatant solution was then decanted off and additional solution was removed with filter candles. The soils were then successively washed with 3-liter portions of distilled water until, in a final washing, the solutions contained less than 10 milliequivalents per liter of chloride ion. This washing required from 15 to 18 liters of water. To each of the 4-kg. portions of soil, from which all possible solution had been removed by the filter candles, 1 liter of Hoagland's nutrient solution was added. This solution contained 5, 5, 2, and 1 millimoles, respec-

tively, of calcium nitrate ($Ca(NO_3)_2$), potassium nitrate (KNO_3), magnesium sulfate ($MgSO_4$), and monopotassium phosphate (KH_2PO_4). The soils, still in the original pans, were then set in the sun until they had dried. After they had been passed through a 2 mm. screen, they were wet to approximately the moisture equivalent with distilled water and frozen at −10° C. for 3 days and again air-dried and screened. Freezing left the soils reasonably friable. The dried aggregates of the Na soil, though easily broken, were harder than those of the washed or Ca soils. The soils as thus treated were used for the wilting coefficient and ultimate wilting point measurements.

Replaceable sodium concentrations in the Na soils were measured after the soils had been removed from the wilting coefficient cans. Total sodium was determined by the uranyl zinc acetate method in an aliquot of the ammonium acetate extract (see section headed Exchange Capacity, p. 405) from which organic matter and silica had been removed. From the total sodium so determined there was subtracted the sodium in solution in the soils as wet with water to three times the moisture equivalents found for the Ca soils. Aliquots of solution for these latter determinations were obtained by centrifuging four 50-gm. portions of each soil in glass tubes. It has been shown by Eaton and Sokoloff (*8*) that the apparent adsorbed sodium, as calculated by subtracting the sodium of aqueous extracts from the total obtained by ammonium acetate extraction, decreases as the soil-water ratio used for extraction is increased.[8] A number of the Na soils of this series as loosely placed in centrifuge cups and wet from below (the standard procedure) absorbed approximately three times the moisture equivalent of water of the Ca soils.

Electrical-conductivity measurements made on the foregoing centrifuge extracts of five of these soils gave values between 63×10^{-5} and 154×10^{-5} reciprocal ohms at 25° C., indicating the presence of between 6 and 15 milliequivalents of salt per liter of solution at this moisture content. This salt included the residual soluble material remaining after the sodium treatments, that of the nutrient solution which was added, and any salt brought into solution during the growth of the sunflowers, minus the salts taken up by the plants.

TABLE 2.—*Exchange capacity and exchange sodium in Na soils*

Soil No.	Exchange capacity per 100 gm.	Sodium in NH_4Ac extract per 100 gm.	Sodium in 3× moisture equivalent extract per 100 gm.	Exchange sodium	
				In soil per 100 gm.	Proportion of exchange capacity
	Milli-equivalents	*Milli-equivalents*	*Milli-equivalents*	*Milli-equivalents*	*Percent*
279	3.34	3.03	0.59	2.44	73.1
1	4.82	3.71	.69	3.02	62.7
4	7.13	5.33	1.37	3.96	55.5
2	8.54	5.54	1.10	4.44	52.0
6	8.02	6.07	1.19	4.88	60.8
3	9.32	7.12	.96	6.16	66.1
9	14.05	10.25	1.38	8.87	63.1
5	16.46	10.42	2.25	8.17	49.6
10	15.83	13.85	1.59	12.26	77.4
8	14.82	9.95	2.16	7.79	52.6
7	20.00	14.47	1.72	12.75	63.8
278	14.77	3.52	1.21	2.31	15.6

[8] Since this paper was written, Kelley (*11*) has published data confirming these findings of Eaton and Sokoloff. His data show for an Imperial Valley soil 5.1 m. e. of absorbed sodium per 100 gm. as determined on the basis of displaced solution, but only 0.6 m. e. on the basis of water extracts. The corresponding values for his Fresno soil 887 were 3.3 and 2.6 m. e., respectively.

As shown by table 2, the sodium chloride treatments did not saturate the soils with sodium and they were better suited, for this reason, to the purposes of the experiment. Neither Gedroiz (*9*) nor Ratner (*13*) was successful in growing plants in soils containing somewhat higher percentages of sodium. In all probability calcium was the principal base in the washed soils, since replacement proceeds in the calcium direction when soils with calcium carbonate are repeatedly treated with distilled water. The sequence of changes accompanying progressive leaching with distilled water of calcareous soils containing sodium (*8*) are (1) dilution of the aqueous phase and (2) a resultant replacement of some of the adsorbed sodium by calcium. As the process is continued, sodium of the aqueous phase is removed by leaching and new calcium comes into solution from calcium compounds, followed by further exchange of calcium for adsorbed sodium.

Soils Treated With Calcium, Magnesium, Potassium, and Sodium

The set of soils treated with calcium, magnesium, potassium, and sodium was given a uniform pretreatment with ammonium acetate and ammonium hydroxide to remove or reduce calcium carbonate and organic matter and thereby make possible a higher percentage saturation with the introduced bases. One-kilogram aliquots of each of the soils were suspended twice in 4 liters of normal ammonium acetate, then in 4 liters of N/10 ammonium hydroxide, and again in 4 liters of normal ammonium acetate. During each suspension the soils were recurrently stirred during the day (in the final ammonium acetate for 4 days), allowed to settle overnight, and then the supernatant solution was decanted and additional solution removed with filter candles. The foregoing treatments did not remove all calcium but none of the final solutions contained more than 10 milliequivalents per liter. The soils were dried, ground to pass a 2-mm. sieve, and divided into five 200-gm. aliquots, only four of which were used.

Each 200-gm. aliquot of each soil was treated three successive times with 800 cc. of normal salt solution ($CaCl_2$, $MgCl_2$, KCl, or NaCl, respectively), and then washed with distilled water, by suspension and stirring and the removal of solution with filter candles, until the chloride content of the suspending solution was less than 5 milliequivalents per liter. The potassium-treated and sodium-treated soils became highly dispersed during the washing with water and certain of them liberated some additional organic matter, but by the end of the washing little came into solution. The soils were finally air-dried, ground, and thoroughly mixed by rolling.

EXPERIMENTAL RESULTS

Effect of Starting Acceleration on the Moisture Equivalent of Calcium-Treated and Sodium-Treated Soils

Although the effect of rate of acceleration on moisture equivalent values has been recognized by others as a factor (*21*), experimental data bearing on it have not been found in the literature. The measurements reported in table 3 show higher moisture equivalent values when the drum is brought to full speed in 15 seconds than when full speed is attained in 4.5 minutes. The effect is especially marked only in the instance of Na soils 7 and 10. Relatively slower accelerations have been most extensively used in the past; otherwise a rapid accel-

eration with the resulting higher values would seem to be desirable, since a closer packing of soil particles is more representative of field conditions.

TABLE 3.—*Effect of starting acceleration on the moisture equivalent of Ca and Na soil*

Time to full speed	Moisture equivalent of soil receiving indicated treatment							
	Soil No. 6		Soil No. 10		Soil No. 5		Soil No. 7	
	Ca	Na	Ca	Na	Ca	Na	Ca	Na
15 seconds	13.2	23.2	21.5	43.0	22.0	37.5	22.2	57.0
4.5 minutes	12.8	21.8	20.7	32.4	20.5	35.6	21.2	51.6

MIGRATION AND SEGREGATION OF SAND, SILT, AND CLAY IN DISPERSED SOILS DURING CENTRIFUGING

Two characteristics of certain of the centrifuged Na soils attracted attention at the outset of this investigation. At the end of the 30 minutes in the centrifuge, free water was present on the surface of the finer-textured Na soils 3, 5, 7, 8, 9, and 10. After these soils were dried the surfaces were glazed, and many times an upper layer curled away from the underlying soil (fig. 1), indicating a segregation of fine particles.

Examinations indicated that the upper layer was composed almost wholly of clay particles. Beneath this clay, silt particles tended to predominate. In soil 7, there was a clearly demarked light-colored silt surface and sand particles were observed to have accumulated in the outer soil near the filter paper. These effects will receive further consideration in the section that follows.

DISTRIBUTION OF MOISTURE IN CALCIUM-TREATED AND SODIUM-TREATED SOILS AFTER CENTRIFUGING

Measurements were made of the distribution of water after centrifuging in five of the Na soils and, for comparison, in the corresponding Ca soils by the following procedure. The original screens were removed from a number of the centrifuge cups and others substituted that could be slipped into place and readily removed. The soils were prepared for the moisture-equivalent determination in the usual way, by weighing 30 gm. of air-dry soil into the cups, allowing them to stand for 24 hours in a tray of water, and gently adding water to the surface of any that had not taken up enough water to wet the surface within half an hour. After the soils had drained for 30 minutes, the cups were placed in the centrifuge drum without jarring and centrifuged for 30 minutes. The cups were taken from the drum, and if there was free water on the surface they were inverted for a few minutes and the inside walls of the cups wiped with absorbent paper. The screens were then carefully slipped off the bottom of the cups and the soils placed on a block of wood slightly concave along the center line and a little smaller than the inside of the cup. By carefully pulling the cup downward, it was possible to expose portions of soil of the desired thickness above the upper edges of the cups. Successive layers, roughly 2 mm. thick, were in turn sliced off with a knife and transferred to weighing cans. Four layers were taken from

each of the Na soils and three from each of the Ca soils. The bottom section of the Ca soils exceeded 2 mm. in thickness, but there was not sufficient for a full 2-mm. fourth layer. These methods, though not very exact with respect to the thickness of the soil layers, were regarded

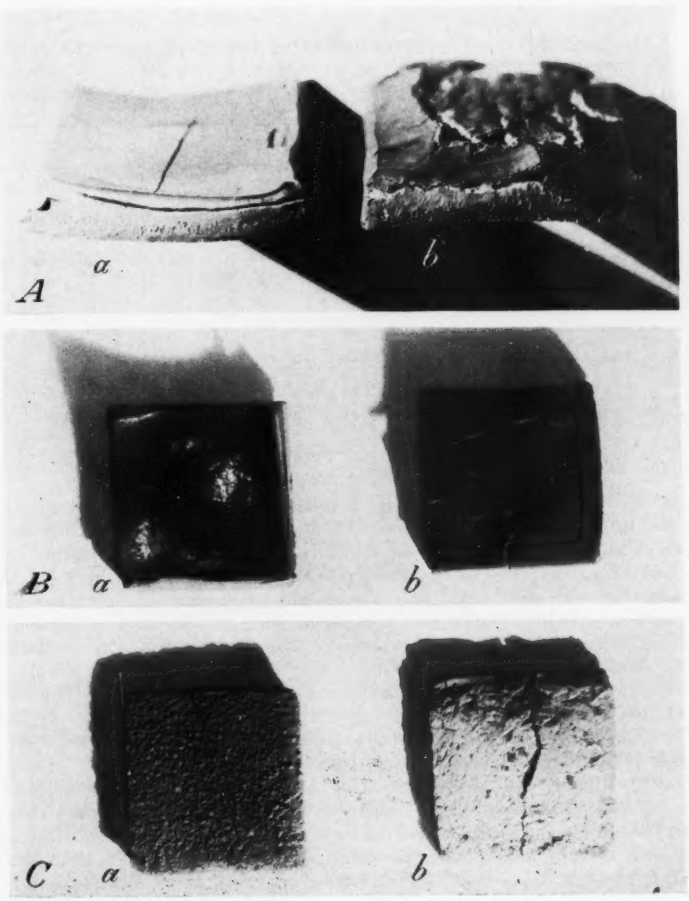

FIGURE 1.—Appearance of centrifuged Ca- and Na-treated soils after drying: *A* and *B*, (*a*) Na soil 10 and (*b*) Na soil 7 after centrifuging and drying; *C*, (*a*) Ca soil 10, (*b*) Na soil 7 with clay layer removed, exposing white calcareous silt layer.

as nonetheless suited to the purpose. Difficulty from crumbling, such as was experienced with Ca soil 5, would have been encountered had the measurements been undertaken with the coarser-textured Na soils.

In conformity with the findings of Veihmeyer, Israelsen, and Conrad (*22*), who worked with untreated soils, it was found (table 4)

that the percentage of water in the Ca soils increased from the inner surface to the outer. The water distribution in each of the Na soils stands in contrast. With these the gradient was reversed. In three of the five Na soils over twice as much water was present in the inner layer as in the outer.

TABLE 4.—*Effect of sodium on distribution of moisture in centrifuged samples*

Soil No.	Moisture content of successive indicated layers of soil from inner surface outward toward periphery of centrifuge in—								
	Ca soils				Na soils				
	0–2 millimeters	2–4 millimeters	Last layer	Weighted mean	0–2 millimeters	2–4 millimeters	4–6 millimeters	Last layer	Weighted mean
	Percent	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*
5	(1)	(1)	(1)		48.4	34.4	29.6	27.8	35.1
					53.8	38.4	32.1	28.4	36.2
					49.0	39.0	34.2	29.9	35.5
7	19.8	21.5	22.7	20.8	99.5	54.3	48.0	40.4	55.0
					94.4	55.7	50.9	44.2	55.5
					76.0	57.5	55.0	47.9	57.8
8	21.5	22.1	23.2	22.2	52.3	39.1	34.3	31.6	38.2
	21.2	22.1	22.2	21.8	52.1	39.1	34.3	31.5	38.1
9	16.3	16.4	17.1	16.6	63.3	24.9	22.2	22.0	30.8
	16.8	17.0	17.3	17.0					
10	20.0	20.2	20.5	20.2	78.4	33.9	29.5	27.2	38.3
	20.7	20.1	20.9	20.5					

[1] Crumbled.

In considering these effects it is necessary to take into account one of the important characteristics of the moisture-equivalent determination. The centrifugal force of 1,000 times gravity is applied not alone to the water, but also to the soil particles. As placed in the centrifugal drum, Na soils contain much water and being dispersed an excellent condition is provided in the plastic masses of the fine-textured soils for particle segregation. The large particles of Na soils, in accordance with Stokes' law, tend to migrate toward the periphery, displacing small particles toward the axis of rotation. The mass of the finer-textured Na soils being somewhat impermeable, a part of the water, which has a specific gravity below that of hydrated clays, is displaced toward the axis of rotation and collects on top of the clay. Accompanying this inward displacement of water there is a further sorting of clay and silt particles by size and specific gravity, giving rise to segregations such as are illustrated in figure 1. Continued centrifuging of Na soils beyond the 30-minute period resulted in some further loss of water, but the rate of movement of water out of the soil was so slow as to eliminate the possibility of removing surface water by any reasonable period of centrifuging.

EFFECT OF ADSORBED SODIUM ON THE MOISTURE EQUIVALENT OF THE 12 SOILS

The moisture equivalents of each of the 12 Ca and Na soils are shown in table 5. In this table there are also shown the clay content and exchange capacity of these soils and the adsorbed sodium of the Na soils.

The moisture equivalents of soils 279, Gold Ridge sandy loam, and 278, Aiken clay, were decreased by treatment with sodium. Soil 279

is highly siliceous, contains little clay, and has a low exchange capacity. Soil 278 is of basic igneous origin, and although it has an apparent clay content of 37.4 percent it possesses few clay properties. The volume weight of this soil is only 1.09, and the ultimate soil particles are irregular in shape, porous, and easily broken. It seems probable that the indicated high clay content may be more apparent than real, since in the mechanical analysis the settling rates of many of the larger fine particles would be slow enough to cause them to be drawn off as clay. Anderson (*1*) found the moisture equivalent of the colloidal fraction of a lateritic soil to be unaffected by sodium treatment.

TABLE 5.—*Effect of adsorbed sodium on the moisture equivalent*

Soil No.	Moisture equivalent			Clay less than 0.002 mm.	Exchange capacity per 100 gm.	Adsorbed sodium per 100 gm.	
	Ca soil	Na soil	Increase in Na soil over Ca soil				
				Percent	*Milli-equivalents*	*Milli-equivalents*	*Percent of total*
279	10.2	9.4		6.5	3.34	2.44	73.1
1	9.2	10.2	1.0	5.9	4.82	3.04	62.7
4	11.2	15.5	4.3	9.2	7.13	3.96	55.5
2	11.9	16.6	4.7	9.6	8.54	4.44	52.0
6	13.3	23.2	9.9	9.1	8.02	4.88	60.8
3	14.5	[1] 33.0	18.5	12.0	9.32	6.16	66.1
9	18.6	[1] 34.0	15.4	25.7	14.05	8.87	63.1
5	21.9	[1] 37.5	15.6	18.4	16.46	8.17	49.6
10	21.4	[1] 42.9	21.5	30.1	15.83	12.26	77.4
8	22.6	[1] 43.7	21.1	21.0	14.82	7.79	52.6
7	22.1	[1] 57.4	35.3	27.3	20.00	12.75	63.8
278	33.0	30.1		26.6	14.77	2.31	15.6

[1] Free water on soil after centrifuging.

The soils are listed (278 excepted) in the order of increasing Na soil moisture equivalents (table 5). Tending to parallel this order, there are increases in the difference between the Ca soil and Na soil moisture equivalents, in clay content, in exchange capacity, and in adsorbed sodium. A closer parallelism would scarcely be expected when account is taken of the variations in the size of clay particles here grouped as less than 0.002 mm., in the hydration characteristics and exchange capacity of different clays, and the differences in soils with respect to particle segregations in the centrifuge. The conclusion obviously follows that the moisture equivalent of soils reflects not only mechanical composition as determined by standard analysis but also soil structure as influenced by the kind and quantity of the adsorbed cations.

The effect of adsorbed sodium on the moisture equivalent is regarded as being consequent to three coincident factors: Hydration, dispersion, and the segregation of a relatively impervious layer of clay on the surfaces of some of the soils. Hydration contributed to the results both because of the tightly held water and because of such relation as hydration may bear to impermeability. Dispersion increased the free surfaces and thereby the water retentiveness of the soils.

THE MOISTURE EQUIVALENTS OF CALCIUM-TREATED SOIL AND SODIUM-TREATED SOIL MIXTURES

When the Ca and Na soils were mixed in equal proportions by prolonged rolling in a sheet of paper, the resulting moisture-equivalent values tended to approach in magnitude the Ca soil values more nearly than the Na soil values (table 6).

TABLE 6.—*The moisture equivalent of mixtures of calcium- and sodium-treated soils*

Soil No.	Moisture equivalent of indicated percentage mixture of Ca soil and Na soil [1]										
	100/0	90/10	80/20	70/30	60/40	50/50	40/60	30/70	20/80	10/90	0/100
279	10.2					10.0					9.4
1	9.2					10.1					10.2
4	11.2					13.0					15.5
2	11.9					12.7					16.6
6	13.3	13.2	13.8	13.3	14.6	15.1	15.6	17.7	18.8	20.4	23.2
3	14.5					17.2					[2] 33.0
9	18.6					22.2					34.0
5	21.9	21.3	21.8	22.5	22.5	23.4	25.2	26.9	30.2	[2] 36.2	[2] 37.5
10	21.4	21.6	21.7	22.3	24.5	[2] 27.2	[2] 30.5	[2] 33.3	[2] 37.7	[2] 41.3	[2] 42.9
8	22.6					27.0					[2] 43.7
7	22.1	21.9	22.8	23.9	27.0	30.6	[2] 38.4	[2] 41.0	[2] 45.5	[2] 54.8	[2] 57.4
278	33.0					31.6					30.1

[1] Numerator = Ca soil; denominator = Na soil.
[2] Free water on soil after centrifuging.

The fact that sigmoid moisture equivalent curves result when Ca and Na soils are mixed in the successive proportions shown in figure 2 would indicate that the relation between adsorbed sodium and either or both hydration and dispersion is not a linear one. Some of Ratner's data (*13*) on the dispersion of clay suspensions by adsorbed sodium also yield a graph with slightly sigmoid characteristics. Bodman and Mahmud (*2*) found a straight line relation between the moisture equivalents of successive mixtures of sand and clay. The migration and accumulation of clay and water on the surfaces of the soils shown in figure 1 unquestionably influenced the shape of the curves. In the soil mixtures where calcium greatly exceed sodium there was little dispersion and, without dispersion little migration. The fact that the upper portions of the curves of soils 7, 10, and 5 are flattened is due in part to the free water. The quantity of free water was observed to increase with the proportion of the Na clay in the mixture. The upper part of the curve soil 6 is not flattened and this soil had no free water on the surface of any of the samples.

The first evidence of a significant increase in the moisture equivalent of soil 6 occurred in the 60/40 mixture, corresponding to 1.9 milliequivalents of adsorbed sodium per 100 gm. of soil. The moisture equivalents of soils 5 and 10 appear to have been increased slightly by the substitution of 30 percent of Na soil, corresponding respectively to 2.4 and 3.6 milliequivalents of adsorbed sodium. The moisture equivalent of soil 7 was increased by the substitution of 20 percent of the Na soil, corresponding to 2.5 milliequivalents of adsorbed sodium. It seems from these limited data that the moisture equivalents of soils are not measurably affected by less than 2 milliequivalents of adsorbed sodium.

The maximum effect on the moisture equivalent was approached in soils 5, 10, and 7 with, respectively, 8.17, 12.26, and 12.75 milliequivalents of adsorbed sodium, corresponding to 49.6, 77.4, and 63.8

percent of sodium in the exchange complex. The maximum effect on the moisture equivalent of soil 6 was not reached or apparently approached with the 4.9 milliequivalent of adsorbed sodium in the 0/100 mixture.

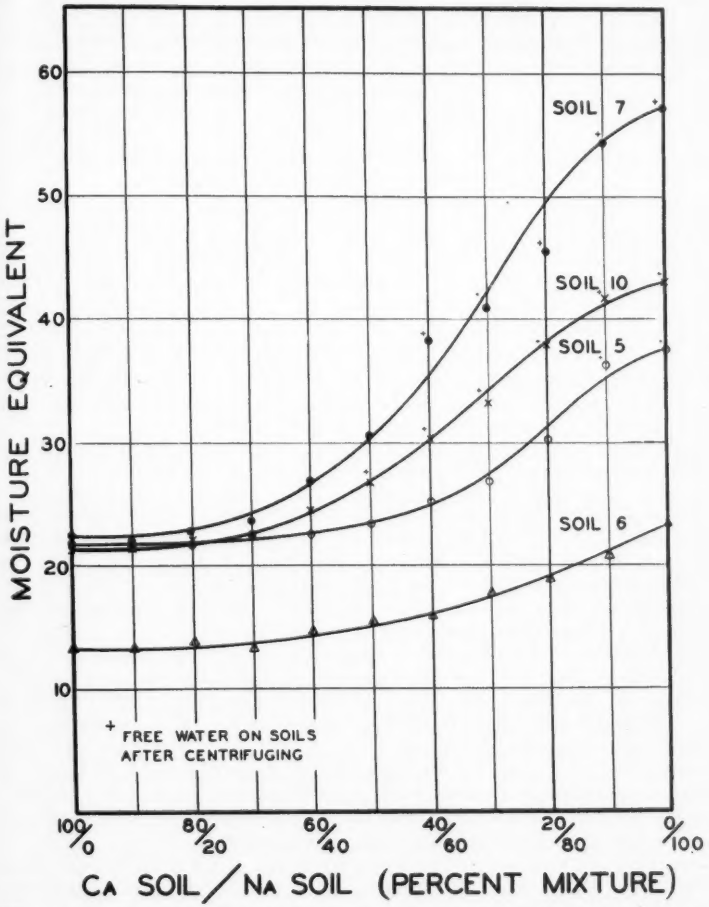

FIGURE 2.—The moisture equivalents of various mixtures of different soils that had been treated with calcium and sodium.

THE MOISTURE EQUIVALENT OF SODIUM-TREATED SOILS WET WITH STRONG ELECTROLYTES

Soil colloids, irrespective of the character of the adsorbed bases, are flocculated when suspended in solutions of strong electrolytes. It follows that adsorbed sodium should be without effect on the moisture equivalent when samples are wet with a strong salt solution rather than with distilled water. The results of measurements made on each of the 12 sodium-treated soils as wet with normal calcium

chloride, and where sufficient soil was available, with normal sodium chloride, are reported in table 7. It is to be observed that the moisture equivalents of the Na soils wet with these solutions tended to be equal to, or slightly lower than, the moisture equivalents of Ca soils wet with distilled water.

TABLE 7.—*Moisture equivalents of Na soils wet with normal $CaCl_2$ and NaCl solutions*

Soil No.	Moisture equivalent of—			
	Na soil wet with normal—		Soil wet with distilled water in usual manner	
	$CaCl_2$	NaCl	Ca soil	Na soil
279	10.2	9.4	10.2	9.4
1	10.0	9.8	9.2	10.2
4	11.6	10.2	11.2	15.5
2	10.9	12.0	11.9	16.6
6	13.6	13.1	13.3	23.2
3	14.2	14.2	14.5	33.0
9	18.5	18.2	18.6	34.0
5	[1] 20.7		21.9	37.5
10	21.4	21.1	21.4	42.9
8	22.9	22.6	22.6	43.7
7	20.9		22.1	57.4
278	29.7	29.6	33.0	30.1
Average	17.0		17.5	29.5

[1] Determined on two 10-gm. samples (values, 26.6 and 25.9 percent) and interpolated after Veihmeyer, Israelsen, and Conrad (*22, table 1, fig. 1*) to 30-gm.-basis.

It is suggested by these results that the moisture equivalent of field soils if determined comparatively (1) as wet with distilled water and (2) as wet with normal $CaCl_2$ or NaCl should provide an index to sodium-induced dispersion and hydration and by inference to reduced permeability from these causes. If saline, the moisture equivalents of Na and Ca soils as taken from the field should be similar. A third and companion measurement is accordingly necessary for a practical utilization of the moisture equivalent as an index to adsorbed sodium, namely, the moisture equivalent of the soil determined after most of the readily soluble electrolytes have been removed by washing and extraction with filter candles. The moisture equivalent so used should provide a practical index, both to the presence of substantial amounts of adsorbed sodium in field soils and to the consequent changes in the permeability characteristics that would be expected to accompany reclamation with nonsaline waters.

The similarity of the moisture equivalents of Ca and Na soils wet with strong electrolytes would seem to eliminate the possibility that the effect of sodium on the moisture equivalent of soils should be attributed to puddling or the mechanics of handling prior to the measurements.

An observable redistribution of soil separates did not occur when the Na soils were wet with strong electrolytes, indicating that the electrolytes increased the attractive forces between particles sufficiently to check migration and the displacement of light particles toward the axis of rotation. The surfaces of the Na samples wet with either of the normal salt solutions resembled the surfaces of the Ca samples, and in no case was free water present on the surface after centrifuging.

The laboratory measurement of the permeability of soils, even on a relative basis, has proved to be a difficult problem in many hands, but some promise of progress is afforded by other measurements wherein soils were leached with solutions of the same composition as their own displaced solutions. The use of such solutions minimizes base exchange and the associated effects on dispersion during successive leachings. By using this procedure creditable agreements have resulted between quadruplicate samples, and the hour-to-hour changes in rate of percolation so often characterizing other permeability measurements have been greatly reduced.

EFFECTS OF CALCIUM, MAGNESIUM, POTASSIUM, AND SODIUM ON THE MOISTURE EQUIVALENT AND HYGROSCOPICITY

The comparative effects of Ca, Mg, K, and Na on the moisture equivalent of soils have been studied only by Anderson (*1*), and he confined his work to the colloidal fraction. It seemed highly probable that Anderson's results might be applied to soils in general, but the significance of the relationships was regarded as sufficiently important to justify independent measurements. The question involved that ties in most closely with irrigation considerations was whether there was ample reason for regarding calcium and magnesium as alike in their physical effects on soils and, if so, whether such potassium as is found in irrigation waters should be classed with the calcium and magnesium or included with sodium in the calculation of the sodium-total base ratio (percent sodium).

The treatments employed in the preparation of the soils used for these experiments, as described earlier in this paper, were more drastic than those used in preparing the Ca and Na soils. Extensive predigestions were made with ammonium acetate and ammonium hydroxide, which were designed to reduce all soils to a common ammonium base and to remove calcium carbonate and humus, but these digestions were not carried to the ultimate end points. Calcium was found in the final ammonium acetate extractions of all soils and humus appeared in some of the first washings of the K and Na soils. Presumably all of these soils approached complete saturation with the respective bases, but no measurements were made. Distilled water washing was continued until the chloride concentrations of the final suspensions were less than 5 milliequivalents per liter.

The moisture equivalents of these soils are compared in two ways, (*1*) when wet with distilled water and (2) when wet with 0.02 normal chloride solutions of the bases corresponding to those adsorbed. The averages of triplicate moisture equivalent measurements are reported except in occasional instances when one of the three values was out of line with the other two.

The moisture equivalents of Ca and Mg soils are found to be similar (table 8) and nearly always less than the K soil. The effect of the sodium ion in this experiment, as in the preceding one, is outstanding, a number of the Na soils yielding moisture equivalent values twice as great as the Ca soils. This is essentially the effect which Anderson (*1*) obtained, but in his series, as in this one, there was one colloid, in addition to the laterite, upon which sodium had little or no effect.

In the presence of 0.02 normal electrolyte, the moisture equivalents of the Ca and K soils were reduced a little, but a consistent effect is not shown in the Mg series. The effect of the 0.02 electrolyte on the

Na soils is marked, but the values are nearer to the Na soil values than to the Ca soil values. From the standpoint of considerations related to the effects of irrigation waters on soils, it seems appropriate to group calcium, magnesium, and potassium ions together and to differentiate these from the sodium ion.

TABLE 8.—*Moisture equivalents of Ca, Mg, K, and Na soils wet with distilled water and 0.02 normal electrolyte*

Soil No.	Moisture equivalent of soil—							
	Wet with water				Wet with 0.02 normal electrolyte			
	Ca soil	Mg soil	K soil	Na soil	Ca soil	Mg soil	K soil	Na soil
279	9.7	10.6	9.7	10.5	9.7	10.7	9.2	10.0
1	9.4	9.8	10.1	10.6	8.9	9.3	9.2	10.8
4	11.7	11.0	11.1	15.7	11.3	11.3	10.3	15.4
2	10.9	10.5	11.0	15.5	10.6	11.3	10.3	15.2
6	12.7	12.9	13.0	18.8	12.2	12.8	11.8	16.4
3	14.0	14.2	15.2	23.6	13.2	14.2	13.9	20.6
9	19.4	19.9	20.1	[1] 33.7	19.1	20.1	18.5	[1] 30.7
5	20.8	22.0	23.7	[1] 45.0	21.1	21.4	22.8	30.4
10	23.9	23.0	26.6	[1] 52.3	23.9	22.6	24.3	[1] 47.4
8	23.3	23.8	26.6	[1] 62.5	22.6	23.8	25.2	39.5
7	22.9	23.6	24.4	[1] 85.3	21.8	23.3	22.6	[1] 54.8
278	36.1	36.1	34.6	32.4	35.2	35.2	32.7	32.6
Average	17.9	18.1	18.8	33.8	17.5	18.0	17.6	27.0
Average effect of electrolyte					−.4	−.1	−1.2	−6.8

[1] Free water on soil after centrifuging.

The influence of the kind of adsorbed base on the hygroscopicity of soils was investigated as an incidental feature of the inquiry by the following procedure. The three oven-dried moisture-equivalent samples of each of the soils of the experiment reported in table 8 were ground and mixed. From each of these, two 25-gm. samples were weighed out and placed on 4-inch watch glasses and exposed on open shelves in a closed concrete basement vault for 15 days. The atmosphere in this vault was humidified by a fan directed into a group of wet towels suspended from a vessel of water. The same fan maintained a good circulation of air throughout the room and over the soils. The average temperature in this vault, as recorded by a thermograph during the period of the experiment, was 20.3° C. (during the 15-day period the maximum was 21.1° and the minimum 18.8°). The average relative humidity during the first 7 days was about 72 percent and during the last 8 days it was 84.1 percent (lowest 81.0 and highest 90.0) as determined twice daily with a sling psychrometer. The partial pressure of water in an atmosphere of 84 percent relative humidity at 20.3° C. is 1.9 cm. Each soil was stirred daily and left ridged to give as great a surface as possible. Starting on the 12th day, selected samples were weighed twice daily until the 15th day by which time the variations were negligible and the gains equaled the losses. The samples were then placed in bottles, and their moisture content was determined by weighing them before and after oven-drying at 105°–110° C. In considering these data, account should be taken of the fact that they are wetting data in contrast with the drying curves represented by the wilting coefficient and ultimate wilting-point results presented in figure 3.

TABLE 9.—*Hygroscopicity of oven-dried Ca, Mg, K, and Na soils when in equilibrium with an atmosphere of 84 percent relative humidity at 20.3° C.*

Soil No.	Hygroscopicity of oven-dried soil—							
	Previously wet with distilled water				Previously wet with 0.02 normal electrolyte			
	Ca soil	Mg soil	K soil	Na soil	Ca soil	Mg soil	K soil	Na soil
	Percent	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*
279	0.93	1.02	0.81	0.87	0.94	1.14	0.93	0.86
1	.90	1.04	.81	.92	.99	1.07	.80	.98
4	1.63	1.65	1.28	1.30	1.65	1.73	1.17	1.54
2	1.75	1.81	1.27	1.46	1.78	1.91	1.35	1.77
6	2.24	2.14	1.63	1.95	2.15	2.23	1.66	1.97
3	2.41	2.31	1.78	2.22	2.34	2.42	1.91	2.24
9	3.75	3.90	2.98	3.15	3.87	4.18	3.02	3.75
5	4.23	4.37	3.15	3.90	4.27	4.52	3.23	3.94
10	4.74	4.70	3.80	4.53	4.98	4.94	3.97	4.85
8	3.94	4.12	3.13	4.20	4.24	4.16	3.20	4.25
7	5.18	5.42	3.81	4.67	5.29	5.62	3.73	4.76
278	7.42	8.06	7.51	7.86	7.50	8.14	7.82	7.57
Average	3.26	3.38	2.66	3.09	3.33	3.51	2.73	3.21
Average effect of electrolyte					+.07	+.13	+.07	+.12

At the end of the absorption period the theoretical concentration of electrolytes in the moisture held by the soils previously treated with 0.02 normal salt solutions is represented by the relation: Moisture equivalent÷hygroscopic moisture × 0.02. Solution concentrations are indicated as high as 232 milliequivalents per liter in Na soil 279 and as low as 82 milliequivalents in Ca soil 7.

The electrolyte increased hygroscopicity in nearly every comparison (table 9), and its average effect, though small, was nearly the same for each of the four bases with which the soils had been treated.

Calcium-treated soils, in keeping with the findings of Anderson (*1*) and with those of Thomas (*19*) at the latter's higher vapor pressures, adsorbed more moisture than did sodium-treated soils. Except for a few determinations (table 9), the magnesium values are slightly higher than the calcium values. Potassium is the low member of the series. Na soils adsorbed less moisture than did Ca or Mg soils but more than K soils. The order both with and without electrolyte is K<Na<Ca<Mg. These results will be referred to again in the final section.

AVAILABILITY OF SOIL MOISTURE IN CALCIUM-TREATED AND SODIUM-TREATED SOILS

The wilting-coefficient and ultimate wilting-point measurements (tables 10 and 11) show that the moisture in Na soils is less available to plants than that in Ca soils, and in nearly all instances the differences are substantial. The differences between the Ca soils and Na soils decrease as the moisture content is reduced from the wilting coefficient to the ultimate wilting point.

The wilting coefficients and ultimate wilting points of soils 278 and 279 were not changed appreciably by the sodium treatments, and these soils, which adsorbed little sodium, showed no positive sodium effects in the moisture-equivalent comparisons. Through the remainder of the series there is a strong positive relation between the effect of sodium in increasing the moisture equivalent and in increasing the

TABLE 10.—*Effect of exchange sodium on the wilting coefficient and ultimate wilting point of Ca and Na soils*

WILTING COEFFICIENT (PERCENTAGE OF MOISTURE)

Soil No.	Washed soil in can No.—							Ca soil in can No.—							Na soil in can No.—						
	1	2	3	4	5	6	Average	1	2	3	4	5	6	Average	1	2	3	4	5	6	Average
279	3.8	3.6	3.3	3.4	3.5	3.8	3.6	4.0	3.4	3.7	3.4	3.3	3.9	3.6	-----	4.3	4.3	3.8	3.9	4.9	4.2
1	3.5	3.5	3.3	3.5	3.8	3.5	3.5	3.6	3.6	3.7	3.7	3.7	3.6	3.7	3.7	5.2	4.6	5.4	4.4	4.5	4.6
4	5.3	5.0	4.9	5.2	5.2	5.0	5.1	4.4	4.5	4.9	5.0	4.6	4.7	4.7	8.2	-----	10.2	-----	-----	7.2	[1] 8.5
2	6.7	5.1	5.1	4.9	4.8	5.2	5.3	5.8	4.8	5.7	5.6	5.1	5.7	5.5	9.5	7.2	8.1	8.1	8.0	10.1	8.5
6	5.5	5.4	5.6	5.8	5.4	5.4	5.5	5.8	5.5	5.6	5.6	5.9	5.6	5.7	7.0	7.0	7.6	9.0	7.6	8.8	7.8
3	6.0	6.2	6.7	6.5	6.1	5.8	6.2	5.7	5.6	6.2	5.7	5.9	5.8	5.8	9.7	8.1	9.0	11.9	9.3	10.0	9.7
9	10.2	10.0	9.8	10.1	10.1	10.0	10.0	10.4	10.2	10.5	9.8	10.3	10.2	10.2	11.4	12.9	12.5	10.7	11.7	11.6	11.8
5	9.3	10.4	10.1	10.1	9.6	9.8	9.9	9.5	9.5	9.5	9.9	10.0	10.1	9.8	13.8	15.6	14.9	13.6	14.7	15.1	14.6
10	11.9	12.1	13.0	12.3	12.3	12.3	12.3	11.4	12.8	11.7	11.8	12.6	11.5	12.0	14.7	15.8	16.2	16.6	15.9	14.1	15.6
8	10.7	10.3	10.3	10.5	9.7	9.8	10.2	12.7	11.4	10.7	12.1	12.0	11.0	11.7	13.9	15.1	13.7	13.9	12.8	12.6	13.7
7	11.2	11.7	11.2	12.2	12.1	10.9	11.6	11.4	11.2	11.2	11.1	11.9	11.5	11.4	15.9	16.2	15.8	15.8	15.4	15.8	15.8
278	22.2	21.4	22.3	21.5	-----	-----	21.9	21.2	21.6	22.7	22.3	-----	-----	22.0	21.8	21.8	22.9	22.9	-----	-----	22.4
Average	-----	-----	-----	-----	-----	-----	8.8	-----	-----	-----	-----	-----	-----	8.8	-----	-----	-----	-----	-----	-----	11.4

ULTIMATE WILTING POINT (PERCENTAGE OF MOISTURE)

Soil No.	Washed soil in can No.—							Ca soil in can No.—							Na soil in can No.—						
	1	2	3	4	5	6	Average	1	2	3	4	5	6	Average	1	2	3	4	5	6	Average
279	3.4	3.2	3.0	3.0	3.3	3.2	3.2	3.3	3.0	3.1	2.8	2.9	3.4	3.1	-----	3.2	3.2	3.3	3.6	3.4	3.3
1	3.3	3.3	3.1	3.3	3.3	3.5	3.3	3.6	3.5	3.6	3.4	3.6	3.3	3.5	3.6	3.9	4.3	4.2	3.8	4.1	4.0
4	4.6	4.6	4.8	5.0	4.6	4.4	4.7	4.4	4.5	4.6	4.4	4.4	4.7	4.5	5.2	-----	5.6	-----	-----	5.2	[1] 5.3
2	4.9	5.0	4.6	4.8	4.7	4.7	4.8	4.6	4.8	4.8	4.9	4.9	4.8	4.8	6.0	5.9	5.8	6.3	6.6	5.7	6.1
6	5.1	5.2	5.0	5.4	5.1	5.3	5.2	5.3	5.2	5.1	5.1	5.0	5.4	5.2	6.8	6.9	7.4	6.6	6.4	6.5	6.8
3	5.5	6.1	5.6	6.0	6.0	5.6	5.8	5.4	5.6	5.7	5.5	5.5	5.5	5.5	6.6	6.5	7.4	7.2	6.9	7.5	7.0
9	9.2	9.1	9.1	10.0	10.0	10.0	9.6	10.0	10.0	9.7	9.0	10.2	9.7	9.8	11.2	10.4	9.6	9.9	10.6	10.3	10.3
5	8.8	9.8	9.3	9.0	8.9	9.2	9.2	9.0	9.5	8.7	9.2	9.0	9.5	9.2	10.2	11.2	10.1	9.9	12.1	11.7	10.9
10	11.4	11.2	11.6	11.5	11.5	12.0	11.5	10.9	10.8	11.2	11.3	11.1	11.0	11.1	12.9	12.5	13.8	12.8	11.8	12.6	12.7
8	10.0	9.6	10.3	10.1	9.2	9.5	9.8	9.8	10.8	10.3	10.9	9.7	11.0	10.4	11.2	12.8	11.0	12.2	12.1	11.4	11.8
7	10.8	10.8	11.2	11.0	10.8	10.7	10.9	11.1	11.1	10.7	10.7	10.9	11.3	11.0	13.2	14.4	14.1	13.0	12.9	13.3	13.5
278	21.3	20.5	21.2	20.8	-----	-----	21.0	20.6	21.0	22.1	20.9	-----	-----	21.2	21.1	21.3	22.0	21.6	-----	-----	21.5
Average	-----	-----	-----	-----	-----	-----	8.3	-----	-----	-----	-----	-----	-----	8.3	-----	-----	-----	-----	-----	-----	9.4

[1] 3 plants in Na soil 4 died before wilting and the remaining 3 were in poor condition.

wilting coefficient and the ultimate wilting point. The coefficient of correlation between differences at the moisture equivalent and ultimate wilting points of Ca and Na soils for soils numbered 1 to 10 is 0.76 ± 0.09, which can be regarded as an indication of the operation of at least some of the same causal factors in the two moisture zones.

TABLE 11.—*Summary of moisture equivalents, wilting coefficients, and ultimate wilting points of washed, Ca, and Na soils*

Soil No.	Moisture equivalent			Wilting coefficient			Ultimate wilting point		
	Washed soil	Ca soil	Na soil	Washed soil	Ca soil	Na soil	Washed soil	Ca soil	Na soil
	Percent	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*
279	10.1	10.2	9.4	3.6	3.6	4.2	3.2	3.1	3.3
1	10.1	9.2	10.2	3.5	3.7	4.6	3.3	3.5	4.0
4	11.2	11.2	15.5	5.1	4.7	[1] 8.5	4.7	4.5	[1] 5.3
2	11.5	11.9	16.6	5.3	5.5	8.5	4.8	4.8	6.1
6	13.1	13.3	23.2	5.5	5.7	7.8	5.2	5.2	6.8
3	13.9	14.5	33.0	6.2	5.8	9.7	5.8	5.5	7.0
9	18.1	18.6	34.0	10.0	10.2	11.8	9.6	9.8	10.3
5	21.2	21.9	37.5	9.9	9.8	14.6	9.2	9.2	10.9
10	22.2	21.4	42.9	12.3	12.0	15.6	11.5	11.1	12.7
8	23.3	22.6	43.7	10.2	11.7	13.7	9.8	10.4	11.8
7	23.9	22.1	57.4	11.6	11.4	15.8	10.9	11.0	13.5
278	32.9	33.0	30.1	21.9	22.0	22.4	21.0	21.2	21.5
Average	17.6	17.5	29.5	8.8	8.8	11.4	8.3	8.3	9.4
Average of soils 1 to 10, omitting 4	17.5	17.3	33.2	8.3	8.4	11.3	7.8	7.8	9.2

[1] 3 of the 6 original plants of Na soil 4 died before wilting and the remaining 3 on which the average is based were in poor condition.

Before continuing the analysis of the results from the standpoint of the soil, the possibility of differences between the plants grown in Ca soils and those grown in Na soils that might affect their ability to utilize soil moisture should be canvassed. McGeorge and Breazeale (*12*) have considered oxygen deficiency as a factor responsible for the wilting of plants in puddled soils. Some support for the idea that puddling, as ordinarily considered, was not an important factor contributing to the results of the present work is provided by the facts: (1) That whether measured in the dry state, in the moisture equivalent cups after centrifuging, or in the wilting coefficient cans, the volumes of the Na soils were greater than the volumes of the Ca soils; and (2) that when the Na soils were wet with strong electrolytes, the moisture equivalent values were nearly the same as those of the Ca soils.

The distribution and the abundance of roots were examined with considerable care both in representative cans before oven-drying and afterwards as the successive cans were emptied for grinding and mixing. Differences were not found, but no examinations were made of the comparative abundance of root hairs. If there had been fewer roots, or a poorer distribution of roots in the Na soils than in the Ca soils, wilting would be expected to have occurred at a higher moisture content in the Na soils.

It may be assumed that the solutions of the Na soils contained substantially higher concentrations of sodium ion than did the solutions of the Ca soils. It is accordingly possible that physiological distinctions should be drawn between the two sets of plants, but so little is known about sodium effects on tissue structure that discussions cannot

be carried far in this direction. As distinct from the Ca roots grown on Ca soil, roots grown on Na soil may have been more highly hydrated and water uptake by them conceivably could have been impeded, but such an effect has never been demonstrated. In some earlier sand-culture experiments in which plants were grown on nutrient solutions high in calcium chloride and high in sodium chloride, respectively, the plant material from the sodium solutions when placed on a Büchner funnel, after drying and grinding, took up and held relatively large quantities of water and could be leached only with difficulty, whereas the calcium plant material behaved in a normal way. The question of whether sodium affects the uptake and movement of water in living plants must be regarded as speculative and there is no present basis for a conclusion.

CAPILLARY POTENTIAL

The term "availability" as phrased by Richards (*14*) involves two notions, namely, (1) the ability of the plant to absorb and to use water with which it is in contact and (2) the readiness or velocity (*6*, *14*) with which the soil moisture moves in to replace that which has been used by the plant. At any given point in the soil water, the capillary potential is numerically equal to the hydrostatic potential and below saturation the quantity is negative. The tension of the soil water at a given capillary potential may be expressed as the length of the suspended water column necessary to produce that tension. The rates of water movement through a soil for a given potential difference will not be the same in different soils, in the same soil at different capillary potentials, or in the same soils as here differentially treated with calcium and sodium. Furthermore, the moisture at the wilting coefficient cannot be regarded either as being uniformly distributed throughout the soil mass or as being in static equilibrium with the plant at wilting. In the wilting range, the plant-soil system is a dynamic one and water withdrawal is continuous, though at a decreasing rate, to the death point.

The present experiments were so conducted as to yield values for each Ca and Na soil at each of three moisture levels, and to each of these moisture levels it is possible to assign at least an approximate capillary-potential value. Schofield and Botelho daCosta (*16*), from freezing-point-depression measurements of their own and calculations from vapor pressures and seed-absorption measurements of others, have computed the pF value of soils (the logarithm of the equivalent capillary tension expressed in centimeters of water column) at the moisture equivalent and at the wilting coefficient. These values ranged from 2.5 to 3.3 for the moisture equivalent and from 4.02 to 4.40 for the wilting coefficient. From the foregoing data, the writers elected to assume an intermediate value of 3.0 for the moisture equivalent and to take Schofield's average value, 4.24, for the pF value at the wilting coefficient. The problem remained of obtaining a suitable value for the ultimate wilting point. By relating the average change in moisture content between the wilting coefficient and the ultimate wilting point of the writers' calcium soils to the corresponding average changes in pF values for like moisture changes in the Schofield and DaCosta Botelho graphs, a pF increment of about 0.16 was indicated. If this is added to the pF value of the wilting coefficient, approximately 4.40 is obtained for the pF value of the ultimate wilting point. For

the purposes here served it is not necessary to assume that pF 3.0 represents the best moisture equivalent value for any soil or for the writers' soils as differentially treated with calcium and sodium. The moisture content values of certain Na soils that have been centrifuged by standard moisture equivalent procedures are not representative of the moisture tension conditions corresponding to a pF value of 3. If other pF values had been used for the moisture equivalent, the wilting coefficient, or the ultimate wilting point, the slopes of the curves (fig. 3) would have been altered but not their general characteristics.

Figure 3 shows moisture percentage (dry basis) plotted against pF for Ca and Na soils: soil 6, a sandy loam; soil 7, a clay; and the aver-

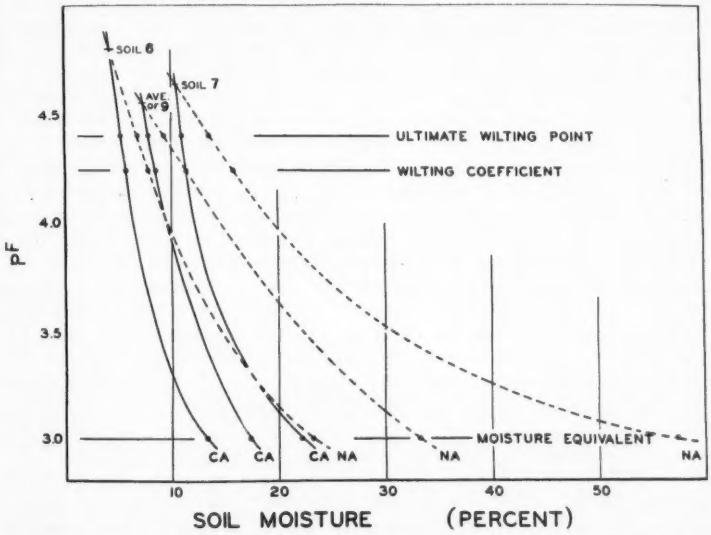

FIGURE 3.—Soil-moisture and pF values of different soils that had been treated with calcium and sodium.

age of the nine soils that showed sodium effects on the moisture equivalent, excluding soil 4, the wilting data for which were not regarded as very accurate. The Ca and Na graphs of corresponding soils are widely separated at the moisture equivalent, but approach each other as the moisture content is decreased, and intersect at a moisture content somewhat lower than the permanent wilting percentage.

From the fact that the Ca and Na graphs intersect, it follows that Ca soils should have higher hygroscopic coefficients than the Na soils in atmospheres substantially below saturation. This finding is in accord with the writers' results (table 9) and those of others.

In keeping with these measurements, it is to be observed that Thomas' (*19*) vapor pressure curves of Ca- and Na-treated soil materials intersected; his Na-treated materials having had a higher moisture-absorbing capacity than his Ca-treated materials from very moist atmospheres and lower from the drier atmospheres. Freezing-point data in a paper by Schofield (*15*) on the penetration of diffuse

double layers may likewise be construed as anticipating the findings of the writers. Those data when extrapolated into the lower moisture range beyond that over which freezing-point measurements are possible also indicate that water is more closely held by an Na soil than by a Ca soil. If extended yet farther the two curves would intersect.

MOISTURE EQUIVALENT-WILTING COEFFICIENT RATIOS

Conclusions with respect to the effect of sodium on the ratio of moisture equivalent to wilting coefficient are made difficult by the fact that free water on the surface of many of these soils after centrifuging caused the observed values to exceed what might be regarded as the true values. For Ca soils 1, 2, and 4 these ratios are 2.5, 2.2, and 2.4, respectively, and for the same Na soils 2.2, 2.0, and 1.8. This suggests that sodium tended to narrow the ratios by raising the wilting coefficient relatively more than the moisture equivalent.

Treatment with sodium increased neither the moisture equivalents nor the wilting coefficients of soils 279 and 278. These soils are of interest, however, for other reasons. The Gold Ridge sandy loam, 279 untreated, has a moisture equivalent-wilting coefficient ratio of 3.0, whereas the Aiken clay has a ratio of 1.6. In the opinion of the writers much of this divergence can be accounted for on the basis of the properties of the ultimate soil particles. The Gold Ridge soil is highly siliceous and the faces of most of the soil particles tend to be smooth, indicating that a large part of the water retained by the soil is exterior to the crystalline surface. A sample of Exeter sandy loam supplied by J. C. Johnston from near Visalia, Calif., had a moisture equivalent nearly four times as great as its wilting coefficient. The siliceous crystalline character and smooth faces of the particles of this soil were more outstanding than in the Gold Ridge soil. The particles of the lateritic Aiken clay, on the other hand, are highly porous and much water is retained, both against gravitational force and plant uptake, as closely held interstitial water, i. e., water within capillary pores. It is obvious that interstitial water retained by a soil both at the moisture equivalent and wilting coefficient would reduce the proportion of moisture-equivalent water available to the plant. The interplanar water of a number of soil colloids represents a third type of retention, but the question of availability of such water to plants and its relation to sodium is obviously complex. Much of it is released to the soil atmosphere as the moisture content of the soil is reduced.

DISCUSSION

Whether the consequences of the state of aggregation or dispersion of the ultimate soil particles are more important in terms of water retention against centrifugal forces and plant uptake than forces involved in the hydration of Ca and Na clays cannot be concluded from these data. Both are probably involved, and the combined soil-water tensions produced are reflected in the water-retention measurements. Dispersion may be consequent to hydration; if so, the two go hand in hand.

The possibility cannot be eliminated that the differences in the availability of moisture in Na soils and Ca soils to plants represent a slower rate of movement through the former soils than through the

latter. In such case it would presumably follow that the soil zones away from the roots were relatively more moist in the Na soils than in the Ca soils, but with equally dry zones in Ca and Na soils at the root surfaces. The distances through which moisture must move to plant roots are relatively great in terms of the thickness of the moisture films at the wilting coefficient. In a soil in which a root or a root hair is situated at intervals of 1 mm., the average distance of water movement is equivalent to 50 times the diameter of a 0.005-mm. silt particle.

If water, in the wilting range, moves primarily by vapor flow, the pertinent distinction between Ca and Na soils in the present connection becomes one of size of capillary pores through which diffusion must take place rather than thickness of water films. Ca soils are characterized by a crumb structure with particle aggregates and voids that are large as compared with those of dispersed Na soils. Vapor diffusion through the Ca soils should accordingly proceed more rapidly than through Na soils.

If moisture movement in the wilting range is primarily movement of film water, it follows likewise that such movement should be slower at a given moisture content in the thinner films covering dispersed particles than in those covering aggregates of closely adhering particles. Water of hydration as such is probably not subject to movement except as it is vaporized into the soil atmosphere.

Irrespective of possible explanations, the finding of immediate significance is that water in dispersed Na soils is less available to plants than that in Ca soils.

SUMMARY

The moisture equivalents of soils partially saturated with sodium were substantially higher than those of the same soils treated with calcium provided most of the soluble electrolyte was removed.

The average moisture equivalents of 12 well-leached Ca, Mg, K, and Na soils were Ca, 17.9; Mg, 18.1; K, 18.8; and Na, 33.8. The average moisture uptake by these soils when exposed (after oven drying) in an atmosphere with a relative humidity of 84 percent at 20.3° C. was K, 2.66; Na, 3.09; Ca, 3.26; and Mg, 3.38 percent

A close parallelism was found for 10 soils between the effect of sodium on the moisture equivalent and the percentage of clay, the exchange capacity, and the quantity of adsorbed sodium.

Adsorbed sodium had negative effects on the moisture equivalent of a soil high in silica and a negative effect on an Aiken clay which is lateritic; both soils had low cation exchange capacities.

The moisture in centrifuged Ca soil samples increased from the inner toward the outer surfaces. An opposite gradient was found in Na soils.

Some migration and segregation of sand, silt, and clay particles occurred when the finer-textured Na soils were centrifuged, a higher proportion of large particles being observed in the outer portions with well-defined clay layers on the inner surface. In certain of the soils water was also displaced toward the axis of rotation and remained on the surface after centrifuging.

Rapid starting acceleration of the centrifuge gave higher moisture-equivalent values than slow acceleration.

Wetting Na soils with normal calcium or sodium chloride solutions gave moisture equivalent values that were nearly the same as those of Ca soils. The moisture equivalent may therefore provide a measure of sodium-induced dispersion of field soils if comparisons are made between leached and unleached soils and soils wetted with a strong electrolyte.

The moisture equivalent of mixtures of Ca and Na soils in successive proportions gave sigmoid graphs. The moisture equivalent was not significantly affected by less than 2 milliequivalents of adsorbed sodium per 100 gm. of soil and values approaching maximum were found when there were 12 milliequivalents or more of adsorbed sodium.

Adsorbed sodium caused soil moisture to be less available to plants. The calcium and sodium averages of nine soils were, at the moisture equivalent, 17.5 and 29.5; at the wilting coefficient, 8.4 and 11.3; and at the ultimate wilting point, 7.8 and 9.2 respectively.

Plotting moisture content against a pF scale, the Ca and Na soil graphs intersect at moisture percentages below the ultimate wilting point. This result confirms earlier work showing lower hygroscopicity of Na soils than of Ca soils in drier atmospheres.

It is suggested that interstitial water held within soil particles against both centrifugal and plant-uptake forces may account for the differences observed in the moisture equivalent-wilting coefficient ratios observed in different soils.

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MODIFICATION OF DIURNAL TRANSPIRATION IN WHEAT BY INFECTIONS OF PUCCINIA TRITICINA[1]

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INTRODUCTION

Earlier investigations (*4*) [3] on the effect of leaf rust (*Puccinia triticina* Eriks.) infection on the water economy of the wheat plant proved that heavy attacks of the disease greatly increased the water requirement of plants of a susceptible variety. It was observed that, while rust-free plants used a greater total amount of water, heavily rusted plants lost more than the controls per unit of dry matter. It also was noted that rusted plants apparently lost water more rapidly in proportion to their surface than rust-free plants. This suggested that leaf rust infection had considerable effect on the transpiration of the host plant and that this factor might lead to severe injury during a shortage of soil moisture. The experiments herein described were designed to measure the transpiration of rusted and rust-free wheat plants and to determine its rhythmic nature.

REVIEW OF LITERATURE

Müller-Thurgau (*7*) studied the effect of various leaf spots on the transpiration of pear, melon, strawberry, and grape, but Blodgett (*1*) was the first to report (1901) on the effect of rust on the transpiration of a host plant. He observed that plants of a species of *Rubus* attacked by *Gymnoconia interstitialis* (Schl.) Lagh. transpired nearly twice as much as rust-free plants. Montemartini (*6*), using cut twigs or shoots bearing rusted leaves placed in water covered with a thin layer of oil, observed that rusts on clematis, violet, rose of sharon, rye, and rose all increased the loss of water by the host plants. He also observed that certain leaf spots increased the loss of water by leaves of several other species of plants. Reed and Cooley (*9*), studying the effect of *Gymnosporangium juniperi-virginianae* Schw. infection on the transpiration of apple leaves concluded that it resulted in lower transpiration. Some diseased leaves were observed to have a water loss only one-fourth as great as comparable healthy leaves.

Apparently the first experiments on the effect of rust infection on transpiration in wheat were conducted by Weaver (*10*), who reported that heavy rust infections greatly accelerated the rate of transpiration. Later, Weiss (*11*) recorded that stem rust infection increased

[1] Received for publication February 28, 1940. Cooperative investigations of the Division of Cereal Crops and Diseases, Bureau of Plant Industry, U. S. Department of Agriculture, and the Kansas Agricultural Experiment Station. Contribution No. 395, Department of Botany and Plant Pathology, Kansas Agricultural Experiment Station.

[2] The writers gratefully acknowledge indebtedness to H. B. Humphrey, of the Division of Cereal Crops and Diseases, for valuable criticisms and suggestions, and to Hugh G. Gauch, graduate assistant, Kansas Agricultural Experiment Station, for assistance during the course of the experiments.

[3] Italic numbers in parentheses refer to Literature Cited, p. 443.

the transpiration of wheat plants but that the differences were not significant for leaf rust.

Nicolas (*8*), using a method similar to that used by Montemartini, found that several rusts increased the transpiration of the host plants from 1.01 to 2.13 times. He also observed that certain smut diseases increased the transpiration of the host. Kourssanow (*5, pp. 350–353*) first reported the effect of smut on the transpiration of a cultivated cereal crop in 1928. He found that wheat plants attacked by *Ustilago tritici* (Pers.) Rostr. transpired 20 percent more than smutfree plants.

Graf-Marin (*3*) studied the effect of mildew caused by *Erysiphe graminis* f. sp. *hordei* on the transpiration of barley plants. He found that diseased plants transpired 14.62 percent more than healthy plants and that the transpiration per unit area was 66.64 percent higher for diseased than for healthy plants. He found also that diseased plants transpired 15 percent more at night than the disease-free control. In searching for the cause of this phenomenon he discovered that the stomata of diseased plants opened wider and that they opened an hour earlier and closed an hour later than those of the controls. He also found that part of the loss of water by mildewed plants was due to the aerial mycelium of the fungus.

Yarwood (*12*) investigated the effect of powdery mildew caused by *Erysiphe polygoni* DC. on the transpiration of excised leaves of red clover under various conditions. The mildewed leaves were found to transpire about the same as healthy leaves during the day but much more than disease-free leaves at night.

Gassner and Goeze (*2*) investigated the effect of infections of *Puccinia glumarum* (Schm.) Eriks. and Henn. on transpiration and assimilation in wheat. They reported in 1936 that heavy rust infection on a susceptible variety greatly increased transpiration and decreased assimilation. The increase was thought to be due to the combined transpiration of the leaf blades and the fungus.

METHODS AND MATERIALS

The experiments were conducted in the greenhouse during the years 1934, 1935, and 1936. Wheat (*Triticum aestivum* L., syn. *T. vulgare* Vill.) plants were grown to maturity in sealed 1-gallon stone jars, each provided with the watering arrangement described in an earlier paper (*4*). A sufficient quantity of good quality garden loam was screened and mixed with a small quantity of fine sand, and the mass was brought to 45 percent of its water-holding capacity. The soil mass was mixed thoroughly and allowed to stand overnight, after which it was again stirred before being used to fill the jars. To facilitate weighing, all jars were brought to equal weight with coarse gravel. The soil was lightly tamped in each jar to render it firm but not hard. It was added a little at a time until there was the same weight of soil in each jar. As soon as the jars had been filled they were sealed with a thin layer of hot paraffin. Five openings were then cut through the seal at equally spaced intervals about 1 inch from the edge of the jar. A clean, well-formed kernel of wheat was pressed into the soil at each opening, and a small quantity of water was added to expedite germination. The jars were then weighed and each was kept at its respective original weight until all seedlings had emerged. At the end of 2 or

3 days, the few seeds that had not germinated were replaced by fresh kernels.

When perfect stands of five seedlings per jar were obtained, all jars were brought to equal weight by the addition of leached sand to the top of the paraffin seal. This effectively sealed the small openings around the plants, any small breaks in the paraffin seal, and the small space that usually developed after a few days between the edge of the paraffin and the jar. Blank control jars set up in this manner and maintained without plants throughout the season proved the method of sealing to be effective.

During the growth of the plants the moisture content of the soil was maintained at approximately the same point by bringing the jars back to their original weight at frequent intervals. During the early stages of growth, watering every second day was found to be sufficient, but later the jars were brought back to constant weight every day. Graf-Marin (*3*) used automatic soil irrigators, stating that daily weighings allowed too much variation in soil moisture, but the writers found the latter method satisfactory.

Two varieties of white spring wheat of Indian origin were used in the experiments of 1934 and 1935, because of their early maturity and strong straw. One of these, Pusa No. 4 (C. I.[4] 8899), is an awnless variety, very susceptible to the physiologic race of leaf rust that was used for inoculum. The other, an awned unnamed hybrid selection of Pusa 52 × Federation (C. I. 11764), is moderately resistant to the same race of rust in the seedling stage and highly resistant in the adult stage. In order to eliminate any differences in the water relations due to the awns of this selection, the hard red spring variety Reward (C. I. 8182) was substituted for it in 1936. This is an awnless selection, having a moderate type of resistance to leaf rust similar to that of Pusa 52 × Federation. It was found to have only slightly more uredia of leaf rust in the heading stage of growth than the unnamed hybrid.

When the plants had reached the desired stages of growth, some of each variety were placed in a large moist chamber, inoculated with a pure culture of leaf rust physiologic race 9, and after 24 hours removed to the greenhouse bench. Inoculation was accomplished by shaking heavily rusted seedlings above the moistened leaves. The controls were moistened and placed in the moist chamber for 24 hours but were not inoculated.

Excellent infection developed on susceptible plants, and much flecking accompanied by a sparse development of uredia appeared on plants of the resistant variety. In the latter there was a moderate development of uredia on the primary leaves in the seedling stage of growth, but at heading time there was only a sparse development of small uredia, accompanied by much flecking. The uninoculated control plants remained rust-free throughout all the experiments, even though they occupied space within a few feet of heavily infected plants.

Rust readings were made on all infected plants when the rust reached full development. Readings were made on each leaf in terms of percentage in conformity with the rust scale used by the Division of Cereal Crops and Diseases, United States Department of Agri-

[4] C. I. refers to accession number of the Division of Cereal Crops and Diseases, formerly Office of Cereal Investigations.

culture. From the many readings obtained, the average percentage infection for each experimental group was determined.

As soon as rust infection was fully developed on the leaves of inoculated plants, all jars were brought to constant weight and were weighed thereafter at regular intervals for a period of several hours to determine the rate of loss of water through transpiration. In 1934, weighings were made every hour, but it was found that changes in weight were more consistent when the weighings were made at 2-hour

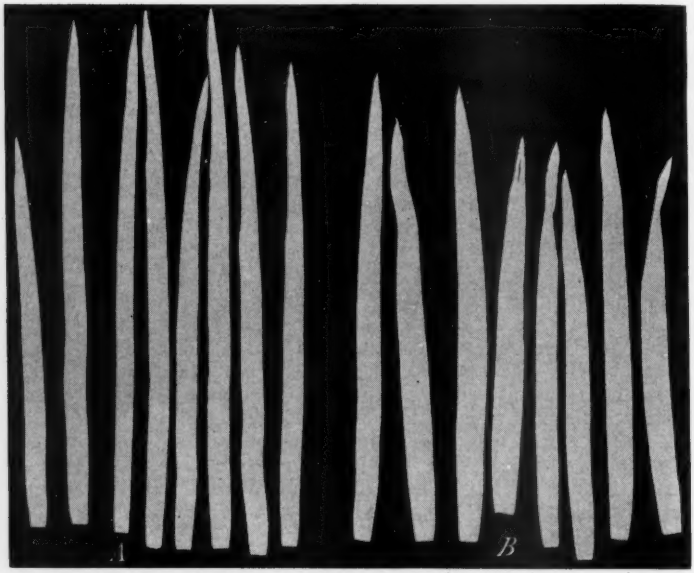

FIGURE 1.—Photograph of blueprint showing differences in size and shape of flag leaves of (*A*) Pusa No. 4 and (*B*) Pusa 52 × Federation spring wheat.

intervals. The latter method was used in 1935 and 1936. With all the jars the weighings were started in the morning and continued throughout the day. The experiments of 1934 were run from 8 a. m. to 9 p. m. on each of 2 consecutive days. In 1935, weighings were made from 8 a. m. to midnight on each of 2 consecutive days, and in 1936 readings were made at 2-hour intervals continuously for a period of 48 hours.

When the weighings were completed, the leaves of all experimental plants were removed and measured. The length and width in millimeters were determined for each leaf. Width measurements were made at the point of greatest width. Blueprints were made of a large number of leaves of each of the varieties, and the areas were determined through the use of a planimeter (fig. 1). The areas thus obtained were compared with those obtained empirically by multiplying the product of two times (length × width) by the factors 0.75, 0.82, and 0.825. Although the factors giving the best fit for each variety varied slightly, it was apparent after many trials that the formula $2(L \times W \times 0.8)$ gave areas very closely approximating those obtained

by planimeter measurements. That formula therefore was used in calculating leaf areas throughout the experiments.

The areas of the stems were determined by multiplying the height (including the head) by the diameter at midheight times 3.1416. The figures thus obtained probably were not entirely accurate because of the inclusion of the heads, which obviously have a greater area than the same length of stems. However, it is known that most wheat stems taper toward the base and toward the head, having the greatest diameter at midheight. Taking diameter readings at that point, therefore, compensated partly for the greater area of the heads.

In calculating the area of transpiring surface, each jar was considered a unit. The sums of the areas of all leaves and stems of the five plants in each jar were calculated and reduced to terms of square meters. By using the weights of water lost during the experimental periods and the transpiring areas of the plants, therefore, it was possible to calculate the water lost per unit of area.

EXPERIMENTAL RESULTS

EFFECT OF LEAF RUST INFECTION ON TRANSPIRATION OF PLANTS OF DIFFERENT AGE

Preliminary experiments were conducted during the greenhouse season of 1933–34 to ascertain the methods most useful for later experiments. Two jars of each variety of wheat were inoculated with leaf rust at each of four stages of growth, viz, the seedling, jointing, booting, and flowering stages. Two similar jars were retained as controls at each stage of growth. Each jar contained five plants, and the four units of each experimental group were selected for similarity of water losses on the basis of their behavior previous to the time of hourly readings.

When leaf rust infection was fully developed on the inoculated plants, the four jars of each variety were brought to their equalized weight early in the morning and weighed hourly thereafter for various periods. The plants tested in the seedling stage were weighed each hour from 9 a. m. to 9 p. m. on January 20 and from 8 a. m. to 4 p. m. on January 21—an elapsed time of 31 hours. The plants studied in the other three stages of growth were weighed hourly from early morning until late evening of 1 day and given a final weighing at 9 a. m. the following morning. The elapsed times for the groups tested in the jointing, booting, and flowering stages were 23, 24, and 23.5 hours, respectively. The amount of water lost through transpiration was recorded at the time of each weighing. The data obtained are presented in summarized form in table 1.

On the basis of the water lost per square meter of transpiring surface, rust infection apparently caused a considerable increase in the transpiration of plants in the seedling, jointing, and heading stages of growth. However, there was only a slight increase in Pusa No. 4 and a slight decrease in Pusa 52 × Federation in the booting stage. The reason for this is not clear, but it probably was due to the small number of experimental units. This is further indicated by the lack of correlation between the amount of rust infection and the percentage increase in the quantity of water lost by infected plants. For example, in the seedling stage of growth, 14.86 percent rust in Pusa No. 4 apparently resulted in an increase of 28.19 percent in transpiration, while only 6 percent infection on Pusa 52 × Federation resulted in a 36.19 percent

increase. In the jointing stage of growth the lack of correlation between amount of rust and loss of water was still more marked. Only in the experiments during flowering did the percentage increase in transpiration seem reasonable for the amount of infection present on the susceptible variety and the absence of infection on the resistant host. There was a moderate development of uredia on the resistant variety in the early stages of growth. However, in later stages infection appeared as copious flecking accompanied by a few very small uredia.

TABLE 1.—*Effect of leaf rust infection on the transpiration of plants of Pusa No. 4 (susceptible) and Pusa 52 × Federation (resistant) wheat at 4 stages of growth in the greenhouse at Manhattan, Kans., 1934*

Stage of growth and variety	Plant group	Experimental period		Average rust infection	Total transpiring area	Total water lost	Water lost per square meter	Increase or decrease in transpiration associated with rust infection
		Dates	Hours					
				Percent	*Square meters*	*Grams*	*Grams*	*Percent*
Seedling stage:								
Pusa No. 4	Rusted	Jan. 20–21	31	14.86	0.561	777	1,385.0	+28.19
	Rust-free			.00	.709	766	1,080.4	
Pusa 52 × Federation	Rusted			6.00	.664	994	1,496.9	+36.19
	Rust-free			.00	.958	1,053	1,099.1	
Jointing stage:								
Pusa No. 4	Rusted	Feb. 5–6	23	40.91	.696	722	1,037.3	+17.94
	Rust-free			.00	.755	664	879.5	
Pusa 52 × Federation	Rusted			1.21	1.027	765	744.9	+17.56
	Rust-free			.00	.955	605	633.5	
Booting stage:								
Pusa No. 4	Rusted	Feb. 13–14	24	32.84	.732	707	965.8	+3.42
	Rust-free			.00	.787	735	933.9	
Pusa 52 × Federation	Rusted			[1]	.914	920	1,006.6	−1.92
	Rust-free			.00	.912	936	1,026.3	
Flowering stage:								
Pusa No. 4	Rusted	Feb. 23–24	23.5	39.76	.642	616	959.5	+20.63
	Rust-free			.00	.621	494	795.5	
Pusa 52 × Federation	Rusted			[1]	.683	524	767.2	+.55
	Rust-free			.00	.726	554	763.0	

[1] Trace.

It was apparent early in the studies that hourly weighings were unsatisfactory because the losses were so small that the error in weighings was likely to be large. It also soon was apparent that plants in the seedling, jointing, and booting stages of growth were not well adapted to an experiment of this kind. The principal reason for this was the impossibility of obtaining rust infection on all leaves. After plants were inoculated, it was necessary to allow about 14 days for the rust to reach full development before readings could be made. During that period the plants put out new leaves and tillers bearing no infection. This is well illustrated by the data in the last column in table 2. The percentages were obtained by dividing the area of rusted leaves by the total area of leaves from each group of inoculated plants.

These data clearly show that a large proportion of the leaves in the seedling and jointing stages of growth had no infection whatever. Furthermore, it was observed that many of the leaves near the bases of the plants during the early stages of growth were only partly active. Many such leaves were brown and shriveled at the tips, and the remaining living portions frequently were chlorotic. Leaves in this condition were extremely difficult to measure accurately.

TABLE 2.—*Comparative number of leaves and percentages of leaf area infected with leaf rust on plants of 2 varieties of wheat inoculated at 4 stages of growth in the greenhouse at Manhattan, Kans., 1934*

Stage of growth and variety	Leaves			Area represented by infected leaves
	Total	Noninfected	Infected	
Seedling stage:	*Number*	*Number*	*Number*	*Percent*
Pusa No. 4	179	48	131	67.05
Pusa 52 × Federation	249	92	157	63.61
Jointing stage:				
Pusa No. 4	205	39	166	84.68
Pusa 52 × Federation	215	86	129	63.86
Booting stage:				
Pusa No. 4	193	28	165	89.68
Pusa 52 × Federation	174	36	138	88.87
Flowering stage:				
Pusa No. 4	106	0	106	100.00
Pusa 52 × Federation	116	0	116	100.00

The results shown in tables 1 and 2, and other observations made during the course of the experiments, indicated that it was impracticable to make weighings oftener than once every 2 hours to determine periodic water losses, and that plants in the flowering stage of growth could be expected to give the most reliable results. It also was apparent that more plants were necessary in each experimental group to decrease the error due to small numbers.

DIURNAL RHYTHM OF TRANSPIRATION IN THE WHEAT PLANT AS AFFECTED BY LEAF RUST INFECTION

One of the most striking features of the preliminary experiments was the sharpness of definition and regularity of the diurnal rhythm of transpiration. Regardless of the age of the plant, the rate of transpiration rose rapidly after 8 a. m. and reached its peak between 2 and 3 p. m. From that point the rate of transpiration declined rather rapidly for a time, then dropped suddenly to a very low point between 5 and 6 p. m. Although the plants were not weighed at intervals throughout the night in the preliminary experiments, the hourly weighings between 5 and 9 p. m. and those made early the following morning showed that the loss of water during the night was very slight as compared with that during midday. For example, nonrusted seedling plants of Pusa No. 4 lost 117.1 gm. of water per square meter of surface between 2 and 3 p. m. and only 9.8 gm. per square meter between 5 and 6 p. m. In the flowering stage of growth, noninfected plants of the same variety transpired 132.0 gm. per square meter between 2 and 3 p. m. and only 27.4 gm. between 5 and 6 p. m. The other variety behaved in a similar manner. It was noted, however, that the time of the sharp drop in transpiration came somewhat later for plants in the flowering stage than for those in the seedling stage. This presumably was due to the difference in the length of day at the time the readings were made. The seedling readings were made on January 20 and the flowering-stage readings were made on February 23, when there was about 45 minutes more daylight at the end of the day.

Although the experiments of 1933–34 were only preliminary in nature, they indicated that leaf rust definitely increased the transpiration of infected plants. There also was strong evidence that

rust infection was accompanied by proportionately greater losses of water at night than during the day, as compared with the rust-free plants. Both of these points are well illustrated in table 3, where the transpiration of rusted and rust-free plants of two varieties of wheat in the juvenile and adult stages of growth are given for successive 2-hour periods from 10 a. m. to 8 p. m. It seems significant that, with but two exceptions, the highest loss of water for both rusted and rust-free plants in both stages of growth occurred during the period from noon to 2 p. m. It also is clear that transpiration was low during the first period in the morning and during the last two periods in the evening.

TABLE 3.—*Loss of water through transpiration by juvenile and adult plants of two varieties of spring wheat at intervals of 2 hours during the day and night in the greenhouse at Manhattan, Kans., 1933–34*

Variety and stage of growth	Plant group	Rust infection	Water per square meter lost through transpiration during successive 2-hour periods ending at—					
			10 a. m.	12 m.	2 p. m.	4 p. m.	6 p. m.	8 p. m.
Pusa No. 4:		*Percent*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*	*Grams*
Juvenile	Rusted	14.86	21.4	185.4	224.6	183.6	48.1	41.0
	Rust-free	.00	28.2	152.3	167.8	173.5	28.2	21.2
Adult	Rusted	39.76	42.1	188.5	224.3	201.0	57.7	67.0
	Rust-free	.00	30.6	183.6	194.8	188.0	48.3	14.5
Pusa 52 × Federation:								
Juvenile	Rusted	6.00	27.1	230.4	224.4	191.3	60.2	34.6
	Rust-free	.00	20.9	154.5	156.6	173.3	47.0	23.0
Adult	Rusted	(1)	8.8	177.2	206.4	174.3	17.5	36.6
	Rust-free	.00	16.5	170.8	209.3	158.3	31.7	50.1

[1] Trace.

The effect of leaf rust infection on the rate of transpiration is shown to best advantage by the susceptible variety Pusa No. 4. In this variety the rusted plants lost more water than the rust-free controls at all readings except that made at 10 a. m. on juvenile plants. Although the readings for both rusted and rust-free plants were low at 8 p. m. as compared with those made at 2 p. m., the percentage increase in transpiration in rusted plants over the rust-free controls was much greater for the night than for the day reading. For example, the transpiration of rusted juvenile plants of Pusa No. 4 at 2 p. m. was 33.85 percent higher than that for the control plants, while at 8 p. m. the rusted plants transpired 93.39 percent more than the controls. In the adult plants of the same variety at flowering time, the rusted plants transpired 15.14 percent more than the controls at 2 p. m. and 362.07 percent more than the controls at 8 p. m.

The difference between day and night increases in transpiration due to rust infection was not so striking in Pusa 52 × Federation. Rusted juvenile plants of that variety transpired 43.29 percent more than the rust-free controls at 2 p. m. and 50.43 percent more than noninfected plants at 8 p. m. It should be kept in mind that the development of uredia on infected plants of this group was only 6.0 percent. The transpiration of the rust-free adult plants was higher than that of the inoculated plants at both the 2 p. m. and 8 p. m. readings. Rust infection at that stage of growth was represented mostly by flecking, with only occasional small uredia.

CUMULATIVE AND PERIODIC TRANSPIRATION OF RUSTED AND RUST-FREE WHEAT PLANTS

The preliminary experiments indicated that leaf rust infection caused an increase in the rate of transpiration of wheat plants but suggested the need for greater numbers of plants and some refinements in technique. During the greenhouse season of 1934–35 the experiments were repeated, with 20 jars each of Pusa No. 4 and Pusa 52 × Federation. Each jar contained 5 plants. No inoculations were made until all plants had reached the flowering stage. At that time plants in 10 jars of each variety were heavily inoculated with leaf rust. The inoculated plants were placed in a moist chamber for 24 hours and then removed to the greenhouse bench. The control plants of each variety received the same treatment as the experimental ones except that they were not inoculated.

When leaf rust infection was fully developed, all dead and partly dead lower leaves, as well as all small weak tillers, were removed from the experimental plants. All jars were brought to original weight at 8 o'clock the following morning. For 48 hours the loss of water through transpiration was recorded by weighings made at intervals of 2 hours except that no readings were made from midnight until 8 a. m. Cumulative losses for the latter period were recorded. The data obtained are shown in table 4 and figures 2 and 3.

TABLE 4.—*Effect of leaf rust infection on the transpiration of plants of Pusa No. 4 and Pusa 52 × Federation wheat during a 48-hour period in the greenhouse at Manhattan, Kans., 1935*

Variety	Plant group	Average [1]				Increase in transpiration due to rust
		Rust infection	Transpiring area	Water lost	Water transpired per square meter	
		Percent	*Square meter*	*Grams*	*Grams*	*Percent*
Pusa No. 4	Rusted	67.01	0.3059	547.2	1,788.8	13.17
	Rust-free	.00	.3193	594.7	1,580.6	
Pusa 52 × Federation	Rusted	2.54	.3282	553.6	1,686.7	5.05
	Rust-free	.00	.3428	550.4	1,605.6	

[1] Per jar of 10 jars containing 5 plants each.

The average loss of water per square meter of transpiring surface in the susceptible Pusa No. 4 was increased 13.17 percent by heavy leaf rust infection, as shown in table 4. At this time the infected plants had an average rust reading of 67.01 percent. The infected plants of Pusa 52 × Federation transpired 5.05 percent more water than the rust-free controls, although the average rust reading of infected plants was only 2.54 percent. A comparison of the increase in transpiration in the two varieties with the rust readings indicates that the increase was proportionately greater in the resistant variety. It must be remembered, however, that the rust readings were based only on the number of uredia present, although there was abundant flecking in addition to the uredia on the leaves and leaf sheaths of the resistant variety. Earlier experiments (*4*) have shown that heavy flecking increases the water requirement of wheat plants, and undoubtedly it also causes an increase in the transpiration.

The total amount of water lost through transpiration during any experimental period is of less interest than the cumulative and periodic water losses, which show when they occur. Figure 2 shows the total cumulative water losses of rusted and rust-free plants of both varieties of wheat studied in 1934–35. During the first test day the total transpiration of the rusted and rust-free groups of Pusa No. 4 was about the same from 8 a. m. to 6 p. m. After the latter hour the rust-free controls transpired very slowly until early the following morning, while the rusted plants continued to lose considerable water,

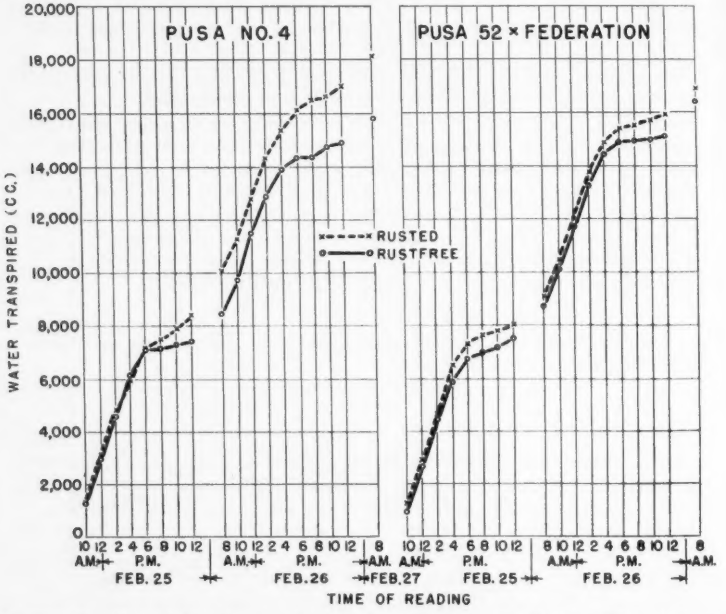

FIGURE 2.—Cumulative losses of water through transpiration by 50 rusted and 50 rust-free plants each of Pusa No. 4 (susceptible) and Pusa 52 × Federation (resistant) during 2 days in 1935.

although the rate was less than that shown during the day. During the second day the results were similar, the rusted plants continuing to transpire more rapidly than rust-free plants after 6 p. m. The trend is shown to be much the same with the resistant variety except that the differences between rusted and rust-free plants are considerably less.

The periodic losses recorded in 1934–35 are shown graphically in figure 3. These show the sharp rise in the rate of transpiration of all plants from early morning to midday and the equally sharp drop after about 4 p. m., regardless of variety or rust infection. The higher rate of transpiration of the rusted as compared with the nonrusted plants of Pusa No. 4 during the night is particularly clear in the readings of February 25. On the following night, except for the 10 p. m. reading, they also transpired more than the rust-free controls for all periods from 6 p. m. to midnight. The small difference

in the transpiration of the rusted and rust-free groups of Pusa 52 × Federation is shown clearly in figure 3 as well as in figure 2.

The experiments of 1934–35 showed rather conclusively that leaf rust infection caused considerable increase in the rate of transpiration of wheat plants of a susceptible variety and to a lesser degree of a resistant variety. They also indicated that one of the principal reasons for the increase was a higher rate of transpiration in rusted plants during the night.

It was desired to repeat the experiments, however, after making a few refinements in procedure. During the greenhouse season of 1935–36, further studies of the problem were made, in which the

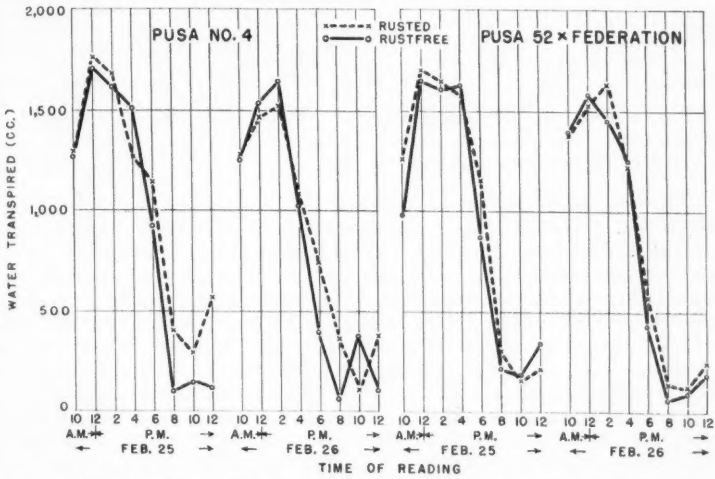

FIGURE 3.—Periodic losses of water through transpiration by 50 rusted and 50 rust-free plants each of Pusa No. 4 (susceptible) and Pusa 52 × Federation (resistant) at 2-hour intervals during 2 days in 1935.

awnless variety Reward was substituted for Pusa 52 × Federation with the thought of eliminating any differences that might have been due to the awns of the latter. The plants were grown in the same containers and handled in the same manner as in the 2 preceding years. At flowering time, 6 jars with Pusa No. 4 and 6 with Reward were selected for the final experiment. These jars were nearly identical in their rate of water loss through transpiration up to the beginning of the experimental period.

When the plants had reached the desired stage of growth, 3 jars (15 plants) of each variety were placed in the moist chamber and heavily inoculated with leaf rust. The 3 control jars of each variety also were placed in the moist chamber but were not inoculated. After rust infection was fully developed, all senescent lower leaves and all but the 3 main tillers on each plant were removed. There remained 5 plants in each jar, each plant having 3 vigorous tillers and 12 active green leaves. The plants were allowed to stand 24 hours after trimming, in order to prevent the recording of any losses due to evaporation from freshly cut surfaces, after which all jars were brought

to their equalized original weights. They were then weighed at the end of each 2-hour period for a continuous 48-hour period. The results are shown in table 5 and figures 4 and 5.

TABLE 5.—*Effect of leaf rust infection on the transpiration of plants of Pusa No. 4 and Reward spring wheat during a 48-hour period in the greenhouse at Manhattan, Kans., 1936*

Variety	Plant group	Average [1]				Increase or decrease in transpiration due to rust
		Rust infection	Transpiring area	Water lost	Water transpired per square meter	
		Percent	*Square meter*	*Grams*	*Grams*	*Percent*
Pusa No. 4	Rusted	57	0.8779	444.3	506.0	+32.7
	Rust-free	0	.9044	345.0	381.4	
Reward	Rusted	1	.8629	312.6	362.2	−1.6
	Rust-free	0	.8555	315.0	368.2	

[1] Per jar of 3 jars containing 5 plants each.

The data presented in table 5 show that heavy leaf rust infection on Pusa No. 4 resulted in an increase of 32.7 percent in transpiration during the 48-hour period. This is nearly three times greater than the increase observed in 1935, although the percentage of rust infection was higher in 1935 than in 1936. However, in 1936 every leaf on the inoculated susceptible plants bore abundant uredia of leaf rust, while in 1935 there were some leaves on all plants that had little or no infection. Also, the difference may be partly due to the smaller number of experimental units used in 1936. This possibility is suggested by the very small decrease in the transpiration of infected plants of the resistant variety (Reward). The important point, however, is that in both 1935 and 1936 heavy leaf rust infection resulted in a considerable increase in the rate of transpiration of the susceptible plants, while that of the resistant plants was affected but little by the rust. These points are graphically illustrated in figures 4 and 5.

Figure 4 shows the total cumulative weight of water lost per square meter of transpiring surface by both varieties during the 48-hour experimental period in 1936. It shows to excellent advantage the gradual increase and the ultimate wide difference in the amount of water transpired between rusted and rust-free plants of Pusa No. 4 as well as the slight difference between similar groups of Reward. It also verifies the observation made in 1935 that the principal increase in the rate of transpiration of rusted over nonrusted plants of a susceptible variety occurs during the night.

The diurnal nature of transpiration in wheat and its modification by leaf rust infection is shown in figure 5. The readings made in 1936 are complete because weighings were made regularly every 2 hours for the entire 48-hour period. Figure 5 shows the transpiration of rusted and rust-free plants of both varieties during each 2-hour period of the experiment. The transpiration of both rusted and rust-free plants rose very rapidly from 8 a. m. until noon, remained high until about 4 p. m., then declined rapidly. The rusted plants of the susceptible variety, except for one reading on January 24, transpired

more at all times than rust-free plants. On the other hand, the control plants of Reward sometimes surpassed the infected plants in

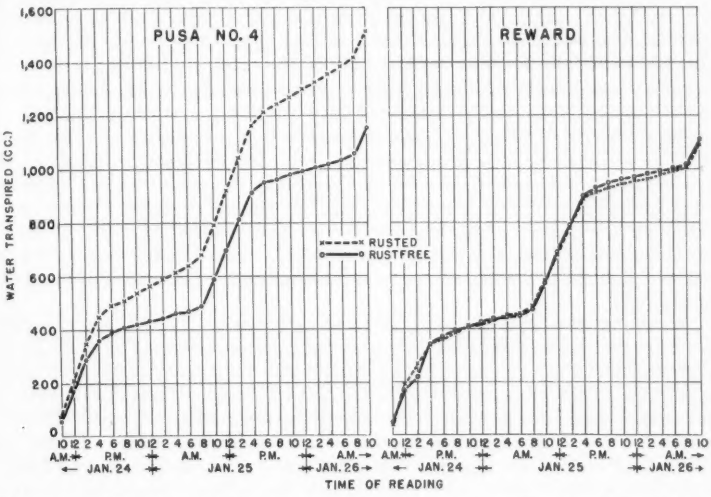

FIGURE 4.—Cumulative losses of water through transpiration by 15 rusted and 15 rust-free adult plants each of Pusa No. 4 (susceptible) and Reward (resistant) spring wheat during a 48-hour period in 1936.

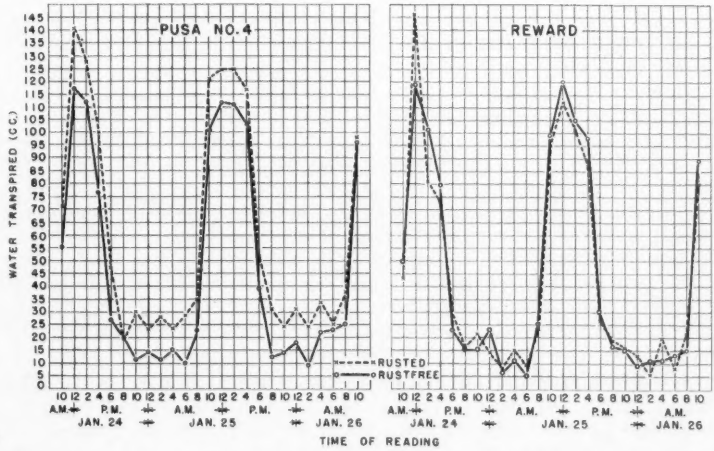

FIGURE 5.—Periodic losses of water through transpiration by 15 rusted and 15 rust-free adult plants each of Pusa No. 4 (susceptible) and Reward (resistant) spring wheat at 2-hour intervals for a 48-hour period in 1936.

transpiration. The fact that the rusted plants of the susceptible Pusa No. 4 transpired more at night than the rust-free controls is shown in both figures 4 and 5.

COMPARATIVE TRANSPIRATION OF RUSTED AND RUST-FREE WHEAT PLANTS DURING PERIODS OF LIGHT AND DARKNESS

Although figures 2, 3, 4, and 5 indicate that the transpiration of rusted wheat plants is higher during the night than that of nonrusted plants, the data in tables 6 and 7 throw further light on the matter. In 1935 and 1936, jars containing rusted and rust-free plants were weighed at intervals for 48 hours. The readings were made during February in 1935 and during January in 1936. At that time of the year darkness begins at approximately 6 p. m. and ends at about 7 a. m. Observations had shown that normal transpiration in the wheat plant began a rapid rise at about 8 a. m. each day and was low and nearly stationary after 6 p. m. It seemed logical, therefore, to consider the period from 8 a. m. to 6 p. m. a period of light, and that from 6 p. m. to 8 a. m. a period of darkness. Thus, there was a 10-hour period of high transpiration and a 14-hour period of low transpiration each day.

It seems desirable for several reasons to present the data obtained for each period of light and darkness rather than to total or average the figures for the 2 days. In the first place, it should be emphasized that the plants were grown in relatively small containers and that no water was added during the 2-day period. The amount of water available for plant use therefore was steadily declining. That fact alone might conceivably affect the results. The data in table 6 show that less water was transpired by both rusted and rust-free plants of both varieties on the second day than on the first day. However, that apparently had no effect on the difference in the rate of transpiration of rusted and rust-free plants.

TABLE 6.—*Average transpiration per jar of 10 rusted and 10 rust-free jars each of 2 varieties of spring wheat during 2 days and 2 nights in the greenhouse at Manhattan, Kans., 1935*

Variety	Plant group	Rust infection	Average transpiring area	Water lost per square meter of transpiring surface			
				8 a. m. to 6 p. m.		6 p. m. to 8 a. m.	
				First day	Second day	First day	Second day
		Percent	*Square meter*	*Grams*	*Grams*	*Grams*	*Grams*
Pusa No. 4	Rusted	67	0.3059	716.34	609.48	260.46	201.96
	Rust-free	0	.3193	704.70	588.12	142.01	147.34
Pusa 52 × Federation	Rusted	3	.3283	732.62	633.84	176.52	144.82
	Rust-free	0	.3428	673.18	609.06	183.99	138.48

Another factor that is known to affect the transpiration of wheat plants in the greenhouse is the variability in greenhouse conditions from day to day. Plants transpire more on bright days, when temperatures are high and the greenhouse ventilators must be open, than on cool, cloudy days when the ventilators are closed. Table 7 shows greater transpiration for the second day in 1936, which was warm and bright, than for the first day, which was cooler and cloudy. This, however, had no influence on the relative rates of transpiration of rusted and rust-free plants. The difference in the transpiration levels in the two seasons was due partly to environmental differences and partly to differences in growth and vigor of plants.

TABLE 7.—*Total transpiration of groups of 15 rusted and 15 rust-free plants each of Pusa No. 4 and Reward spring wheat during day and night periods of 2 consecutive days in the greenhouse at Manhattan, Kans., 1936*

Variety	Plant group	Rust infection	Transpiring area	Water lost per square meter of transpiring surface			
				8 a. m. to 6 p. m.		6 p. m. to 8 a. m.	
				First day	Second day	First day	Second day
		Percent	*Square meter*	*Grams*	*Grams*	*Grams*	*Grams*
Pusa No. 4	Rusted	57	0.8779	486.4	541.0	186.8	206.2
	Rust-free	0	.9044	388.1	464.4	99.5	107.2
Reward	Rusted	1	.8629	372.0	426.5	107.8	100.8
	Rust-free	0	.8555	373.0	453.5	99.3	90.0

Table 6 shows that rusted and rust-free plants of Pusa No. 4 transpired nearly the same during the two daylight periods, but that rusted plants lost much more water than the rust-free controls during the periods of darkness. In the resistant Pusa 52 × Federation, however, there was little difference between rusted and rust-free plants during either of those periods.

The data obtained in 1936, as shown in table 7, corroborate those obtained in 1935. In both years the rusted plants of the susceptible variety transpired nearly twice as much at night as nonrusted plants, while there was little difference in the nighttime transpiration of infected and noninfected plants of the resistant variety.

While the figures presented in tables 6 and 7 show that rusted plants of a susceptible variety transpire much more during the night than do rust-free plants of the same variety, they do not show the difference for comparable time periods. The daylight periods were only 10 hours, while the periods of darkness were 14 hours. The data presented in table 8 therefore were calculated on the basis of the transpiration per square meter per hour. This obviously causes wider differences in transpiration between the periods of daylight and darkness than those shown in tables 6 and 7.

Although only data for 1935 are shown in table 8, the results for 1936 were similar. It will be noted that the difference between the transpiration of rusted and rust-free plants of both varieties was very small during the daylight periods, although rust apparently caused a slight increase in all cases. In the resistant variety the transpiration during the night was much lower than that during the day, but there was not much difference between the rate of transpiration of rusted and rust-free plants as shown by the percentages of increase or decrease.

The most outstanding feature of the data shown in table 8 is the enormous percentage increase in the nocturnal transpiration of rusted plants of Pusa No. 4 over the rust-free controls of that variety. During the first night the rusted group transpired 83.41 percent more than the nonrusted group, but during the second night the increase was only 37.08 percent. The lower percentage probably was due to the smaller amount of water available for transpiration as the plants neared the end of the 48-hour period. For the two periods of darkness the rusted plants of Pusa No. 4 therefore transpired an average of

60.24 percent more per square meter per hour than the rust-free controls. The difference between the rates of transpiration of heavily rusted and rust-free plants at night was still more marked in 1936, when the rusted plants of Pusa No. 4 lost on an average 89.62 percent more water per square meter per hour than did rust-free plants.

TABLE 8.—*Transpiration per square meter per hour in rusted and rust-free plants of a susceptible and a resistant variety of wheat during 2 periods each of daylight and darkness in the greenhouse at Manhattan, Kans., 1935*

Variety and plant group	Transpiration per square meter per hour			
	8 a. m. to 6 p. m.		6 p. m. to 8 a. m.	
	First day	Second day	First day	Second day
Pusa No. 4 (susceptible):	*Grams*	*Grams*	*Grams*	*Grams*
Rusted	716.34	609.48	186.04	144.27
Rust-free	704.70	588.12	101.43	105.24
Increase	*Percent* +1.65	*Percent* +3.63	*Percent* +83.41	*Percent* +37.08
Pusa 52 × Federation (resistant):	*Grams*	*Grams*	*Grams*	*Grams*
Rusted	732.62	633.84	126.09	103.44
Rust-free	673.18	609.06	131.42	98.91
Increase or decrease	*Percent* +8.83	*Percent* +4.07	*Percent* −4.05	*Percent* +4.58

It seems clear from the foregoing data and discussion that the increase in the transpiration of rusted wheat plants of a susceptible variety over that of rust-free plants is very largely due to the higher rate of transpiration of the rusted plants during the night. The reasons for the higher nighttime rate of transpiration of rusted plants are speculative. Graf-Marin (*3*) and Gassner and Goeze (*2*) both suggest that higher nocturnal transpiration was partly due to transpiration of the rust fungus. Graf-Marin worked with a mildew that does not rupture the host-plant tissues as do the rust fungi. He also observed that the disease stimulated stomatal action in the host plant in a way that would favor higher transpiration. The studies reported herein throw little light on the causes of higher nocturnal transpiration in heavily rusted wheat plants. However, the large amount of ruptured plant tissue in such plants strongly suggests that transpiration through the ruptures caused by the uredia may be an important factor. The large amount of fungus tissue exposed in rust uredia also suggests that the fungus itself may be the cause of considerable loss of water. This loss of water by evaporation from thin-walled fungus cells can hardly be considered a function of the host plant but obviously is inseparable from normal transpiration. Two years' observations, therefore, suggested that the higher transpiration of rusted plants during the night was due partly to injury caused by the rupturing of the epidermis of the host plant by rust uredia and partly to transpiration by the fungus itself. That the latter is an important factor is suggested by the fact that the average transpiration of infected resistant plants during the night was only slightly higher than that of the noninfected controls.

The difference between the transpiration of rusted and rust-free plants of both resistant and susceptible varieties during daylight hours

was small. This probably was due to the very high transpiration level during the day. In all instances, however, the average transpiration of infected plants was slightly higher than that of the control plants during the day.

SUMMARY

The effect of leaf rust infection on the transpiration of a resistant and a susceptible variety of spring wheat was studied in the greenhouse during 3 years.

Plants were grown to maturity in sealed stone jars and leaf and stem areas were calculated. Blueprints made of representative wheat leaves proved that the formula $2(L \times W \times 0.8)$ gave leaf areas very near planimeter readings.

Plants in the flowering stage were found to be superior to younger plants or seedlings for studies of this nature.

Normal transpiration in the wheat plant was found to rise very rapidly during the morning hours, reach a maximum about noon, and decline rapidly in the late afternoon, reaching a very low point about 6 p. m. and remaining extremely low during the hours of darkness.

Observations were made on the transpiration of rusted and rust-free plants at 2-hour intervals for 48-hour periods in 1935 and 1936. It was found that rusted plants of the susceptible variety transpired 13.17 percent and 32.7 percent, respectively, more than nonrusted controls for the entire period in the 2 respective years.

The diurnal rhythm of transpiration was seriously disturbed in rusted plants of the susceptible variety. Such plants transpired much more than rust-free controls during the night. In 1935 the increase in nocturnal transpiration of rusted plants over the controls was 83.41 percent, while in 1936 it was 89.62.

The higher rate of transpiration of rusted plants at night apparently was due partly to transpiration through ruptures in the cuticle caused by the uredia and partly to the transpiration of the fungus itself.

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THE INFLUENCE OF THE AWNS UPON THE RATE OF TRANSPIRATION FROM THE HEADS OF WHEAT[1]

By HUGH G. GAUCH,[2] *formerly graduate research assistant, Kansas State College of Agriculture and Applied Science,* and EDWIN C. MILLER, *plant physiologist, Kansas Agricultural Experiment Station*[3]

INTRODUCTION

Although the exact physiological role of awns is not well established, it is generally considered that under certain climatic conditions the awns of wheat are useful structures. On this account awned varieties of wheat have been selected and grown for the most part in regions of limited rainfall. The data which have been obtained relative to the functions of the awns are frequently fragmentary, contradictory, and sometimes the result of questionable methods. Work has been in progress since 1935 at the Kansas Agricultural Experiment Station to try to determine some of the functions of the awns of wheat. The data herein reported, relative to the influence of the awns on the rate of transpiration from the heads, are thus only a portion of those obtained in the general study of the function of the awns of the wheat plant.

REVIEW OF LITERATURE

The quantity and quality of the grain produced has been the measure, for the most part, of the effects of the removal of the awns from cereals. Only a relatively few of the investigators have studied the influence of the awns on the rate of transpiration. Zoebl and Mikosch (*10*)[4] stated that the awns of barley are definite organs for transpiration. The awned heads transpired four to five times more water than the awnless heads. At the time of the functioning of the awns about one-half of the total transpiration of the plant was from the heads. The transpiration of the awns was most intense at the time of the greatest migration of reserve materials into the grain. An unsigned article in a German publication (*1*) stated that the transpiration from barley heads is decreased by the removal of the awns.

Vasilyev (*9*) in Russia studied the function of awns in rye, wheat, and barley. In experiments with Beloturka wheat, he found that the awns transpired 63.3 percent of the water evaporated by the heads, and in another variety 60.3 percent of the total transpiration of the heads was through the awns. The maximum transpiration of the awns was at flowering.

[1] Received for publication December 5, 1939. Contribution No. 396 from the Department of Botany, Kansas Agricultural Experiment Station.

[2] A portion of the thesis submitted by the senior author to the Graduate Council, Kansas State College of Agriculture and Applied Science, in 1937 for partial fulfillment of the requirements for the degree of master of science.

[3] The authors are especially indebted to C. O. Johnston, associate pathologist, U. S. Department of Agriculture, for selecting and supplying the wheat used in this study and for his helpful suggestions and criticisms.

[4] Italic numbers in parentheses refer to Literature Cited, p. 457.

Schmid (*7*) stated that the awns are actively concerned in transpiration, assimilation, and respiration. The de-awning of a wheat plant lowered its transpiration 10 to 30 percent. He found that the awned heads transpired relatively more water by night than by day, while with awnless heads as well as with the lamina of the leaf, the reverse was true. Since the awns and the lamina both have numerous stomata, Schmid considered that the stomata of the lamina opened wider during the day than did those of the awn.

Perlitius (*6*) found that the awned heads of wheat and barley showed a higher transpiration rate than the awnless heads. The same fact was reported by Conti (*2*) for the heads of durum wheat.

Schulze (*8*) showed that awned barley plants transpired much more than those which had been de-awned or were naturally awnless. The peak of transpiration was reached when the grain was in the milk stage.

METHODS OF EXPERIMENTATION

CULTURAL METHODS

The wheat plants were grown in glazed stone jars of 1-gallon capacity with the same amount of soil mixture firmly tamped into each jar. The soil mixture was made by adding 1 part sand to 4 parts of fertile loam soil. Sufficient water was added to this mixture to bring the moisture content to 45 percent of the water-holding capacity of the soil. The method used to facilitate the distribution of water throughout the soil in the jars and to prevent its loss from the surface was the same as that described by Johnston and Miller (*4*).

Before planting, a ½-inch cork borer was heated and used to make five equidistant holes in the paraffin layer above the soil. A germinating grain of wheat, with a radicle one-fourth of an inch long, was planted in the soil beneath each of these holes. The seeds were planted on October 25, 1935, and on November 2, 1936. At regular intervals, varying according to the stage of growth, the jars were weighed and sufficient water was added in each case to bring them to their original weight.

During the experimental work in 1935–36, it was found that a hybrid wheat of Indian origin, Pusa 52 × Federation, because of its type and rapidity of growth, and other characteristics, was the most desirable plant to use. It was used, therefore, exclusively in the experiments of 1937. The general appearance of these plants when they had attained their full vegetative growth is shown in figure 1.

SELECTION AND DE-AWNING OF HEADS

In the experiments of 1935–36 no record was kept of the development of the heads so their exact stage of growth at any given time was not known. Thus during 1935–36, the pairs of heads used in the transpiration experiments were matched as closely as possible only with regard to size. During 1936–37, however, a detailed record of 215 heads was kept so that in all the experimental work for that year the heads would be not only of the same size, but also of a known age and stage of development.

When the awns protruded visibly from the boot, the culm of the head was tagged and detailed notes were taken thereafter at 2-day intervals. Some heads were used for observational purposes only,

and spikelets were removed from these from time to time to determine the progress or rate of grain formation. Thus it was not necessary to remove any spikelets from the heads used in the transpiration studies to determine their stage of development.

FIGURE 1.—Typical jars of Pusa 52 × Federation wheat, showing the condition of the plants on February 8, 1936, when the plants were 75 cm. in height. The planting date was October 25, 1935.

Four apparently similar heads were selected for each experiment. Two heads were used as controls, while the awns were clipped from the other two with small, sharp scissors. The appearance of the awned and de-awned heads at the time of an experiment can be observed in figure 2.

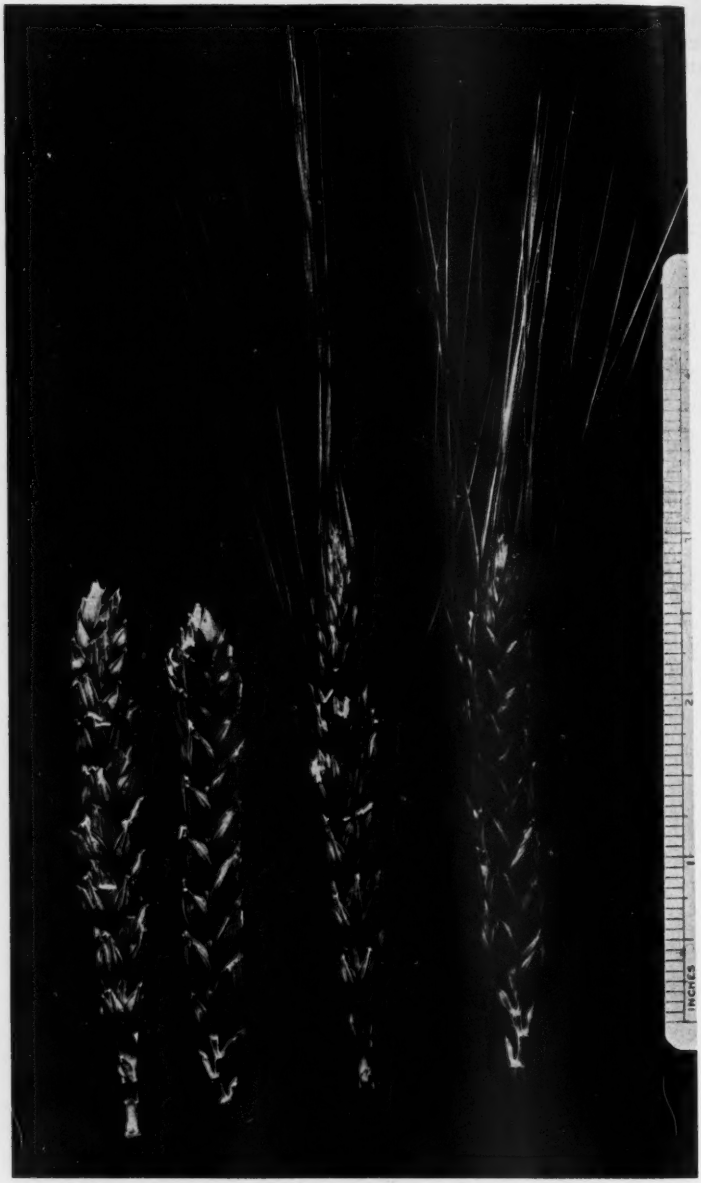

FIGURE 2.—Totally de-awned and intact awned heads of Pusa 52 × Federation wheat.

The objection may be raised that the injury resulting from the removal of the awns might influence transpiration to some extent, and that the lowering of the transpiration of the de-awned heads might be due to this effect and not to the loss of the awns. There is no means of measuring this effect, but after a period of 2 months when the wounds incident to removal had been healed for a long time, the transpiration from the de-awned heads was still as low as, or lower than it was at the beginning of the experiment.

METHOD OF MEASURING TRANSPIRATION

In these experiments, a modification of the Freeman (*3*) method was used in determining the rate of transpiration of the heads. The general procedure is as follows: An aspirator provides the suction that draws the air over the plant parts which are enclosed in a glass chamber and through weighed U-tubes containing a water absorbent. In the present experiments granular calcium chloride was used as the absorbent. The air that passes over the plant parts contains both the moisture released from these parts and that which is normally present in the air. The amount of moisture in the air at the time of the experiment is determined by the use of a control apparatus, figure 3, *h*, *m*, in which the same amount of air as is drawn over the plant parts is drawn through the U-tubes of the control apparatus, and the moisture contained therein is absorbed. The weight of the moisture collected in the control apparatus is then deducted from the amount found in the air which passed over the plant parts. The remainder is the weight of the water given off from the portion of the plant used in the experiment.

The apparatus used in 1936–37 is shown in figure 3. Three units were operated simultaneously in each experiment: One, *m*, a control, for determining the water content of the air, another, *n*, for determining the loss of water from the awned intact heads, and a third, *o*, for determining the water loss from the de-awned heads. In figure 3 the unit for the control, *h*, *e*, *m*, is shown at the extreme left. At the center, *i*, *e*, *n*, and right, *j*, *e*, *o*, are the units which determine the rate of transpiration of two awned heads and two de-awned heads, respectively.

The glass chamber (fig. 4) used was a type of small specimen jar. This chamber was 7 by 2½ inches, outside measurements, and had a capacity of 500 cc. It was fitted with a No. 8 three-holed rubber stopper split into two portions so that the line of cleavage passed through two of the holes to permit the introduction of the peduncles of the heads. In the third hole the outlet tube was inserted. The holes that held the two peduncles were sufficiently large to allow free intake of air around these parts when suction was applied by the aspirator to the outlet tube. The portions of the peduncles that protruded into the glass chamber were coated with vaseline to prevent transpiration, and when they were taken from the chamber this coating was removed to prevent injury to the peduncles.

A double set of the U-tubes was placed in connection for each unit (fig. 3, *e*, center), but only one pair was open during any one experiment. At the end of one experiment, the used pair of U-tubes was closed and a new experiment was started immediately by opening the unused pair of tubes. During the second experiment, the used pair

was removed for weighing and a new set was installed in the system for a later experiment.

Without the use of such a set-up, it would have been necessary to remove the three pairs of U-tubes and to install new ones at the end of each experiment. Under such conditions, the heads must be removed from the glass chambers or other special precautions must be taken to prevent an accumulation or condensation of moisture within these chambers. By the use of the extra pair of U-tubes, as above described, it was possible to make as many as four 1-hour determinations of

FIGURE 3.—The modified Freeman apparatus used in the study of the transpiration of wheat awns: *a*, Large aspirators; *b*, small aspirators; *c*, hose for filling aspirators; *d*, trough for carrying water from aspirators; *e*, U-tubes; *f*, atmometer; *g*, pinch-cocks of aspirators; *h*, glass chamber for control; *i*, glass chamber for awned heads; *j*, glass chamber for de-awned heads; *k*, plants bearing awned heads; *l*, plants bearing de-awned heads; *m*, control unit; *n*, *o*, units used for water determinations on head of wheat.

transpiration without disturbing the heads or pausing between experiments.

Two 18-liter aspirators, *a*, and one 4-liter aspirator, *b*, were used in each of the units. As the water siphoned from the aspirators, it emptied into a trough, *d*, and was carried by it to a nearby drain. The aspirators were refilled by a garden hose attached to the water line, *c*. The small aspirators were also connected into the system leading from the glass chambers and these were run while the heads were being inserted and the control was being set up. This procedure prevented an accumulation or condensation of moisture within the chambers during that time.

When arrangements were completed for beginning the experiment, the three small aspirators were closed, each pair of U-tubes, *e*, in each unit was opened, and one of the 18-liter aspirators, *a*, for each unit

was started. The time of starting the experiment was then recorded and the porous-cup atmometer, *f*, was read. When the first aspirators were empty, the rubber tubes connecting them with the system were clamped with pinchcocks and the second aspirator of each unit was started immediately. While the second aspirators were running, the first ones were refilled, primed, and reconnected to the system so that they could be started again as soon as they were needed. By this arrangement a constant suction could be provided throughout the experiment. In most of the experiments each of the two aspirators of a unit was emptied twice, so that 72 liters of water were used. As the 72 liters of water siphoned from the aspirator, 72 liters of air were necessarily drawn in to replace them. In order to reach the aspirator the air had to enter around the peduncles at the base of the heads (fig. 4), flow upward around the heads, down the outlet tube, and through the weighed U-tubes (fig. 3, *e*) before it could enter the aspirator where the vacuum was created.

Since it took approximately 15 minutes for one aspirator to empty, the experiment had a duration of about 1 hour. By adjusting the screw clamps, *g*, *h*, on the outlet tube of the aspirators, the units could be made to finish at approximately the same time. If the experiment lasted slightly more or less than 1 hour, the transpiration values were reduced to that of an hourly period. At the close of each experiment, the atmometer, *f*, was read and its evaporation likewise calculated to the hourly basis.

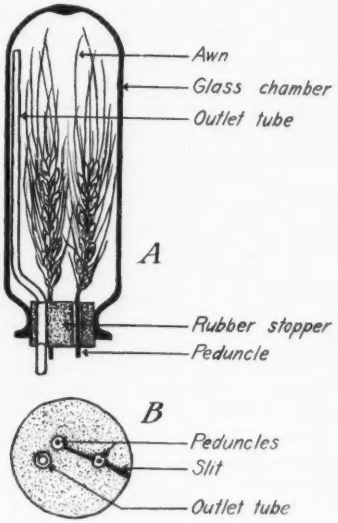

FIGURE 4.—*A*, Diagrammatic drawing of the glass chamber used; *B*, surface view of the rubber stopper which was used in the glass chamber, showing the slit between two holes to permit insertion of the peduncles, and cross sections of the outlet tube and of the peduncles around which the air entered.

At the beginning of a series of experiments, the amount of transpiration from two pairs of intact heads was determined for a given period of time. The ratio of the rate of transpiration of one pair of heads to that of the other was then calculated. In this paper, this ratio is termed the "initial ratio" of transpiration. After this initial ratio had been determined, one pair of heads was de-awned (fig. 2) and its rate compared with that of the intact pair in numerous determinations.

EXPERIMENTAL RESULTS

During 1935–36, fifty-five 1-hour determinations were made to test the effects of the removal of the awns on the transpiration of the head. These experiments were of a preliminary nature, and through them the technique used during 1936–37 was perfected. Although

the results of the experiments performed during the first year agreed in general with those herein reported, they are not tabulated because they were obtained under various methods of technique.

During the year 1936–37, one hundred and eight 1-hour determinations of transpiration exclusive of those used in determining the initial ratio were made on the same variety of wheat as used the first year. Experiments were conducted with seven sets of heads (table 1). The number of determinations on each set ranged from a minimum of 12 to a maximum of 21. The experiments on the sets ranged from 15 to 35 days. Three of the sets began at or before the flowering stage, two at the early milk stage, one at the milk stage, and the seventh at the soft dough stage, and all were continued until the grain was ripe. The detailed data of these seven sets are shown in table 1. The de-awned heads in all except one of the determinations showed a decidedly lowered transpiration in comparison with the intact heads. The one determination that showed an increase in transpiration of the de-awned heads over that of the intact heads occurred in set 6 from 10:30 to 11:29 a. m. on February 20, 1937. In that experiment the ratio of the transpiration of the de-awned heads to that of the awned ones was 1.04. Since in 108 experiments this is the only case in which the transpiration from the de-awned heads exceeded that from the intact ones, it seems probable that some error occurred.

Table 1 shows in detail the data that were obtained in each set of experiments. In this table are found the date of each experiment, the time of day that it was performed, the general stage of development of the head, the nature of the weather, the rate of evaporation from a porous cup atmometer, the temperature, the number of days that had elapsed from the time of de-awning until the experiment, the number of days since flowering, the hourly rate of transpiration from the de-awned heads and the awned heads and the ratio of these two rates of transpiration. Any detailed information relative to these experiments can be obtained by a study of table 1.

Johnston and Miller (*5*) studied the influence of leaf rust (*Puccinia triticina*) upon the rate of transpiration of wheat plants of the same variety as that reported herein and at the same time that these experiments were being conducted. One observation which was made by them at that time and which was not reported in their experiments was that the amount of water transpired from the heads of wheat was relatively small in comparison with the amount lost in the same time from the entire plant. The heads transpired only from 1 to 5 percent of the water lost from the entire plant.

In figure 5, the determinations of transpiration are plotted according to the number of days after flowering. By this method of plotting the data, the amount of transpiration is correlated with the stage of development of a plant or plant part. For the most part, the determinations have been grouped into 3-day units. The data were obtained by averaging the results of all experiments occurring within the time limit of any unit. Thus, in the unit 1 to 3 days after flowering, the values representing the transpiration of awned and de-awned heads are the averages of thirteen 1-hour determinations as taken from the various sets of experiments (table 1).

TABLE 1.—*Transpiration experiments, with 7 sets of awned and de-awned heads of Pusa 52×Federation wheat, in the greenhouse, Manhattan, Kans., 1937*

Set No. and date	Temperature	Time of day [1]	Evaporation from atmometer per hour	Water in each 72 liters of air	Net water loss per hour		Stage of development	Days from time of first flowering	Time after de-awning	Transpiration ratio of de-awned to awned heads	Weather
					2 awned heads	2 de-awned heads					
SET NO. 1	° F.		*Cubic centimeters*	*Milligrams*	*Milligrams*	*Milligrams*		*Number*			
Jan. 19	71	10:40–11:40	1.4	305.5	220.7	222.3	Flowering	0		[2] 1.01	Sunny.
Do	72	3:05– 4:12	1.4	391.3	161.3	113.0	do	0	5 minutes	.71	Cloudy.
Jan. 20	75	9:01–10:10	1.4	459.3	201.6	132.0	do	1	1 day	.65	Do.
Do	74	10:32–11:34	1.3	276.2	825.2	461.4	do	1	do	.56	Sunny.
Do	68	1:42– 2:44	1.1	318.5	376.2	280.5	do	1	do	.75	Cloudy.
Do	68	3:05– 4:05	1.0	448.7	184.4	132.5	do	1	do	.72	Do.
Jan. 21	73	9:20–10:24	1.4	471.3	183.1	155.4	do	2	2 days	.85	Do.
Jan. 22	73	8:17– 9:17	1.5	410.5	195.8	125.3	do	3	3 days	.64	Do.
Jan. 23	68	8:17– 9:21	1.3	428.1	167.1	120.8	do	4	4 days	.72	Sunny.
Jan. 24	78	8:02– 9:01	1.3	532.3	188.8	144.5	do	5	5 days	.77	Do.
Jan. 25	67	8:59– 9:57	.8	507.9	318.6	205.2	do	6	6 days	.64	Do.
Jan. 26	70	8:39– 9:40	1.3	447.0	290.2	172.0	do	7	7 days	.59	Do.
Jan. 27	67	8:04– 9:03	1.0	530.5	178.9	112.0	Between flowering and early milk.	8	8 days	.63	Do.
Do	70	9:03–10:04	1.4	433.5	338.8	188.7	do	8	do	.56	Do.
Do	72	2:59– 3:59	1.9	314.7	359.7	200.8	do	8	do	.56	Do.
Do	72	3:59– 5:00	1.0	446.5	213.5	125.6	do	8	do	.59	Do.
Jan. 28	63	7:55– 8:55	1.0	459.2	102.2	31.6	Early milk	9	9 days	.31	Cloudy.
Feb. 4	65	8:45– 9:44	1.0	406.2	294.8	123.6	Soft dough	16	16 days	.42	Sunny.
Feb. 8	74	12:02– 1:00	2.0	396.6	337.3	170.2	do	20	20 days	.50	Do.
Feb. 15	72	9:59–10:57	1.2	555.1	215.1	132.4	Hard dough	27	27 days	.62	Do.
Feb. 19	67	12:39– 1:37	.8	654.6	94.3	34.6	do	31	31 days	.37	Cloudy.
Feb. 22	75	8:45– 9:44	1.5	495.2	15.4	11.9	Grain ripe	34	34 days	.77	Do.
SET NO. 2											
Jan. 21	74	10:52–11:54	1.4	459.9	247.5	252.7	Flowering	0		[2] 1.02	Do.
Do	75	2:41– 3:41	1.2	494.7	202.2	130.9	do	0	6 minutes	.65	Do.
Do	70	3:41– 4:43	1.1	451.5	178.2	114.6	do	0	1 hour	.64	Do.
Jan. 22	72	9:34–10:35	1.5	393.5	221.6	188.9	do	1	1 day	.85	Sunny.
Jan. 23	68	9:40–10:40	1.4	404.2	314.6	235.9	do	2	2 days	.75	Do.
Jan. 25	73	10:11–11:11	1.5	475.2	349.0	226.7	do	4	4 days	.65	Do.
Jan. 26	73	1:26– 2:26	2.0	322.2	287.5	167.8	do	5	5 days	.58	Do.
Do	73	2:26– 3:27	1.7	309.7	331.2	196.2	do	5	do	.59	Do.
Feb. 4	71	9:57–10:55	2.2	363.7	366.9	238.3	Milk	14	14 days	.65	Do.
Feb. 8	70	1:06– 2:06	1.8	364.3	307.8	187.1	Soft dough	18	18 days	.61	Do.
Feb. 15	73	11:04–12:02	1.8	520.2	279.2	229.4	Hard dough	25	25 days	.82	Cloudy.
Feb. 19	70	1:43– 2:42	.9	646.4	109.1	81.2	do	29	29 days	.74	Do.
Feb. 22	77	9:49–10:49	1.9	391.9	262.0	122.6	Ripe	32	32 days	.47	Sunny.

See footnotes at end of table.

TABLE 1.—*Transpiration experiments, with 7 sets of awned and de-awned heads of Pusa 52×Federation wheat, in the greenhouse, Manhattan, Kans., 1937*—Continued

Set No. and date	Temperature	Time of day	Evaporation from atmometer per hour	Water in each 72 liters of air	Net water loss per hour		Stage of development	Days from time of first flowering	Time after de-awning	Transpiration ratio of de-awned to awned heads	Weather
					2 awned heads	2 de-awned heads					
SET NO. 3	° F.		*Cubic centimeters*	*Milligrams*	*Milligrams*	*Milligrams*		*Number*			
Jan. 22	75	1:30- 2:30	1.5	404.6	374.1	319.9	Flowering	1		[2].86	Sunny.
Do	77	2:30- 3:30	1.6	473.0	451.0	356.7	do	1		[2].79	Do.
Jan. 23	70	1:34- 2:35	1.5	366.0	367.3	199.5	do	2	4 minutes	.54	Do.
Do	68	2:35- 3:35	1.6	346.1	412.2	213.0	do	2	1 hour	.52	Do.
Jan. 24	71	9:20-10:21	1.6	468.7	376.8	216.6	do	3	1 day	.57	Do.
Do	70	11:11-12:11	1.6	380.4	468.2	273.6	do	3	do	.58	Do.
Do	73	12:11- 1:11	2.0	360.7	440.7	293.1	do	3	do	.67	Do.
Jan. 25	67	1:16- 2:15	1.6	280.6	268.5	182.6	do	4	2 days	.68	Do.
Do	67	2:15- 3:17	1.8	262.6	355.4	186.5	do	4	do	.52	Do.
Jan. 26	68	9:53-10:53	1.4	364.2	326.9	216.2	do	5	3 days	.66	Do.
Feb. 4	73	1:26- 2:24	1.8	372.3	307.2	155.4	Milk	14	12 days	.51	Do.
Feb. 8	68	2:13- 3:11	1.6	339.8	375.7	199.8	Soft dough	18	16 days	.53	Do.
Feb. 15	75	1:50- 2:49	1.4	516.7	340.9	165.9	Hard dough	25	23 days	.49	Cloudy.
Feb. 19	70	2:47- 3:46	.9	618.2	122.0	64.8	do	29	27 days	.53	Do.
Feb. 22	73	1:25- 2:24	1.9	311.6	231.7	103.3	Ripe	32	30 days	.46	Sunny.
SET NO. 4											
Jan. 28	67	9:27-10:26	.9	581.0	121.8	122.9	Early milk	9		[2] 1.01	Cloudy.
Do	74	1:04- 2:04	1.4	503.5	221.4	116.1	do	9	4 minutes	.52	Do.
Do	73	2:04- 3:04	1.5	480.7	222.2	117.8	do	9	64 minutes	.53	Do.
Jan. 29	73	8:17- 9:16	1.1	579.9	102.8	66.8	Milk	10	1 day	.65	Do.
Do	68	9:16-10:16	.9	606.9	375.0	67.9	do	10	do	.18	Do.
Do	67	1:27- 2:28	.8	562.4	166.5	116.1	do	10	do	.70	Do.
Do	66	2:28- 3:28	.7	549.9	132.6	76.8	do	10	do	.58	Do.
Jan. 30	67	8:31- 9:30	1.0	536.9	124.3	100.0	do	11	2 days	.80	Do.
Do	68	9:30-10:29	1.1	560.2	161.2	123.2	do	11	do	.76	Do.
Do	73	1:22- 2:21	1.2	634.3	226.9	124.3	do	11	do	.55	Do.
Do	73	2:21- 3:20	1.2	639.4	227.4	125.3	do	11	do	.55	Do.
Feb. 4	73	2:38- 3:37	1.5	402.5	245.1	157.4	Soft dough	16	7 days	.64	Sunny.
Feb. 8	68	3:16- 4:14	1.5	357.3	302.3	191.0	do	20	11 days	.63	Do.
Feb. 15	75	2:54- 4:53	1.4	500.1	246.0	125.5	Hard dough	27	18 days	.51	Cloudy.
Feb. 19	71	3:56- 4:55	.9	641.4	64.9	37.8	do	31	22 days	.58	Do.
Feb. 22	72	2:29- 3:29	2.0	300.8	103.3	28.3	Ripe	34	25 days	.27	Sunny.
SET NO. 5											
Jan. 31	66	8:32- 9:30	.9	553.7	234.8	229.3	Early milk	10		[2].98	Do.
Do	70	9:36-10:36	1.3	555.6	295.8	199.1	do	10	6 minutes	.67	Do.
Do	70	10:36-11:37	1.6	527.4	406.4	250.7	do	10	66 minutes	.62	Do.
Do	72	11:37-12:35	2.0	450.7	393.4	198.7	do	10	2 hours	.51	Do.

Feb. 1	68	9:00- 9:59	1.1	554.4	477.9	275.2	do	11	1 day	.58	Do.
Do	72	11:00-11:59	.9	493.2	522.4	328.5	do	11	do	.63	Do.
Do	72	12:00-12:58	1.9	437.2	498.2	307.3	do	11	do	.62	Do.
Do	72	2:00- 3:00	1.9	364.6	407.1	237.8	do	11	do	.58	Do.
Feb. 2	70	8:00- 8:59	1.2	552.1	203.7	158.4	Milk	12	2 days	.78	Do.
Do	73	10:00-10:58	2.1	435.6	442.3	273.5	do	12	do	.62	Do.
Do	70	1:00- 1:59	1.4	379.1	336.9	219.8	do	12	do	.65	Cloudy.
Do	73	3:00- 4:00	1.3	496.4	403.4	216.7	do	12	do	.54	Do.
Feb. 5	70	9:00- 9:58	1.0	548.2	217.3	123.4	do	15	5 days	.57	Do.
Do	73	9:58-10:57	1.7	458.3	301.9	173.2	do	15	do	.57	Do.
Do	70	3:37- 4:35	1.1	458.3	205.3	141.3	do	15	do	.69	Do.
Do	70	4:35- 5:33	.8	599.5	175.0	101.2	do	15	do	.58	Do.
Feb. 9	68	10:23-11:21	1.0	472.8	203.6	111.0	Soft dough	19	9 days	.55	Do.
Feb. 16	75	11:56-12:53	2.2	276.5	464.7	243.2	Hard dough	26	16 days	.52	Sunny.
Feb. 20	67	9:24-10:23	.9	518.0	140.6	72.6	do	30	20 days	.52	Cloudy.
Feb. 23	73	12:37- 1:36	1.9	389.5	273.6	140.0	Ripe	33	23 days	.51	Sunny.
Set No. 6											
Feb. 6	67	8:44- 9:43	.8	492.2	332.9	277.8	Milk	14		[2].83	Do.
Do	70	9:50-10:49	1.3	458.5	521.4	323.6	do	14	5 minutes	.62	Do.
Do	67	1:42- 2:40	.9	384.5	332.1	204.3	do	14	4 hours	.62	Do.
Do	70	2:40- 3:38	1.2	426.4	505.4	296.8	do	14	5 hours	.59	Do.
Feb. 7	72	8:45- 9:43	1.0	690.5	164.2	123.2	do	15	1 day	.75	Cloudy.
Do	73	9:43-10:44	1.3	629.9	198.1	141.5	do	15	do	.71	Do.
Do	75	1:45- 2:43	1.7	449.9	297.3	189.6	do	15	do	.64	Do.
Do	74	2:43- 3:43	1.2	518.5	268.8	164.5	do	15	do	.61	Do.
Feb. 9	73	1:03- 2:02	1.0	566.3	270.8	178.2	Soft dough	17	3 days	.66	Do.
Do	75	2:03- 3:01	1.2	568.2	350.4	218.9	do	17	do	.62	Sunny.
Do	73	3:01- 4:00	1.9	403.2	397.0	265.6	do	17	do	.67	Do.
Feb. 16	75	1:00- 1:59	2.3	268.5	334.5	226.9	Hard dough	24	10 days	.68	Do.
Feb. 20	68	10:30-11:29	1.1	535.5	131.8	136.8	do	28	12 days	1.04	Cloudy.
Feb. 23	78	1:41- 2:41	2.2	375.9	372.2	240.0	do	31	15 days	.64	Do.
Feb. 26	71	1:45- 2:45	1.3	399.5	166.8	103.7	Ripe	34	18 days	.64	Do.
Set No. 7											
Feb. 12	67	9:18-10:16	.8	607.7	39.8	46.2	Soft dough	20		[2]1.16	Do.
Do	70	10:17-11:21	1.5	446.7	251.4	199.0	do	20	2 minutes	.79	Do.
Do	73	3:05- 4:03	1.4	426.6	187.6	137.5	do	20	5 hours	.73	Do.
Do	72	4:03- 5:02	1.3	451.5	192.4	133.1	do	20	6 hours	.69	Do.
Feb. 13	72	2:45- 3:43	2.4	245.0	464.2	295.6	do	21	1 day	.64	Sunny.
Do	68	3:43- 4:43	1.7	253.5	367.4	244.5	do	21	do	.67	Do.
Do	70	4:43- 5:42	1.1	450.3	219.1	151.7	do	21	do	.69	Do.
Feb. 14	66	8:05- 9:04	.7	619.3	174.3	118.4	Hard dough	22	2 days	.68	Do.
Do	70	9:04-10:03	1.4	530.3	351.8	225.1	do	22	do	.64	Do.
Feb. 16	75	2:05- 3:04	3.8	264.5	409.1	252.4	do	24	4 days	.62	Do.
Do	75	3:04- 4:03	1.7	338.3	346.3	193.3	do	24	do	.56	Do.
Feb. 20	65	3:25- 4:25	1.0	468.7	217.2	120.7	do	28	8 days	.56	Cloudy.
Do	68	4:25- 4:55	1.2	497.6	171.6	124.4	do	28	do	.72	Do.
Feb. 23	73	2:46- 3:45	2.2	369.9	265.0	140.8	do	31	11 days	.53	Do.
Do	71	2:50- 3:51	1.4	348.4	165.6	83.4	Ripe	34	14 days	.50	Do.

[1] All experiments were conducted during daylight hours.
[2] Initial ratio.

Figure 5 shows that the amount of transpiration rose rapidly to a maximum during the period of 1 to 3 days after flowering, following which there was a relatively sudden decline until about the eighth day after flowering. The rate of transpiration then rose and maintained approximately a constant level until the eighteenth day. The rate then declined until the twenty-first day, when it again increased to a second marked maximum approximately 26 days after blooming. During the next 3 days the rate declined to the second lowest point for the season, then increased during the next 3-day period, and again declined to the minimum.

The first maximum is in accordance with the observations of Vasilyev (*9*) that the maximum transpiration by the awns of Beloturka

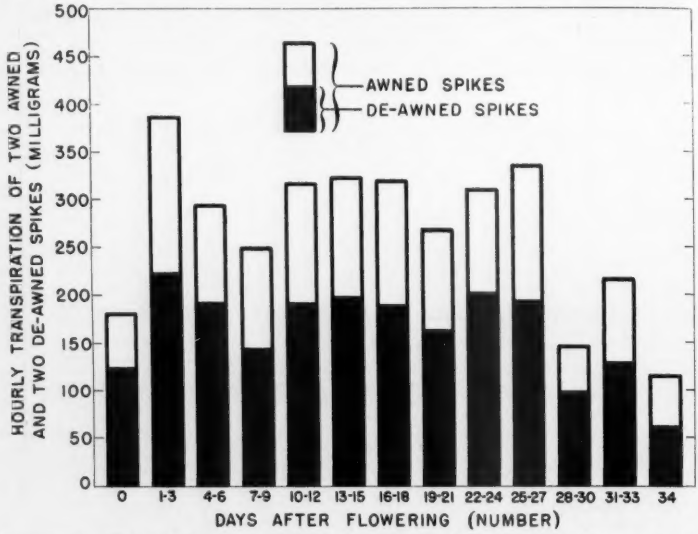

FIGURE 5.—The total transpiration of awned and de-awned heads of wheat at 3-day intervals from flowering to maturity. The transpiration of the awned heads is represented by the total height of the column and includes both the light and shaded portions; the transpiration of the two de-awned heads is represented by the height of the shaded portion of the column.

wheat was at the time of flowering. That portion of the diagram which represents the rate of transpiration from the eighth to the twenty-ninth day after flowering corresponds to the curve reported by Perlitius (*6*) for spring wheat. For winter wheat, Perlitius obtained a double curve with the maxima occurring at the onset of fruit formation and just preceding the milk stage. Since he made determinations at only six periods between heading and maturity, he did not obtain sufficient data to indicate whether other fluctuations occurred before or after periods of observation.

In the present experiments it is clearly indicated that the variations in the rate of transpiration of the heads were not correlated with the atmospheric conditions of the environment as shown by the rate of evaporation from the porous-cup atmometer. It also seems evident

that there was no correlation between variations in light conditions and variation in the transpiration of the heads. The curve of transpiration of the heads during their development and maturity show two very evident maxima. It seems evident that the heads of wheat at flowering and at the approach of maturity of the grain show marked maxima in the rate of transpiration. The significance of this change of the transpiration rate in the metabolism of the plant is not known.

SUMMARY

The effects of de-awning upon the rate of transpiration of the heads of Pusa 52 × Federation wheat were studied in the greenhouse in 1935–36 and 1936–37. During the first year fifty-five 1-hour determinations were made, and the technique for the following year was developed. During the second year one hundred and eight 1-hour determinations were made.

The rate of transpiration of the awns was determined by the Freeman method, which was modified and improved to meet the specific requirements of the experiment. By this method the net transpiration of the awned and de-awned heads was determined quantitatively. The amount of transpiration attributable to the awns was thus obtained.

The rate of transpiration of the awned and de-awned heads was determined at various times from just before flowering until the head had fully matured. The average ratio of the amount of water transpired from the seven sets of intact heads that later were de-awned to the seven sets of heads that served as controls during the experiments was 0.976. In 108 experiments made as early as 2 minutes after de-awning and as late as 34 days thereafter, and from the early flowering stage of the heads to complete ripeness of the grain, the average ratio of the amount of water transpired from the de-awned to the awned heads was 0.61. On a percentage basis the de-awned heads transpired 38.9 percent less than did the awned heads.

The curves of transpiration of the awned and de-awned heads paralleled each other throughout the experiments from flowering to maturity. These curves showed a maximum rate of transpiration of the heads at flowering and another as the maturity of the head was approached. These variations in the rate of transpiration seemed to be independent of the environmental conditions as indicated by evaporation from a porous-cup atmometer, but the cause of these variations is not known.

The rate of transpiration from the heads of wheat is evidently decreased by de-awning, but the significance of transpiration from these parts in the metabolism of the plant is unknown.

Although the awns of wheat are active in the transpiration of the head, they do not ordinarily transpire more than 1 to 5 percent of the total amount of water lost from the plant.

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A DEVELOPMENTAL ANALYSIS OF KOHLRABI AND CABBAGE STEMS [1]

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INTRODUCTION

Many horticultural and agronomic problems are concerned, either directly or indirectly, with the subject of the relative growth rates of the cells, tissues, and organs of plants. The rate of cell division in relation to rate of enlargement is a fundamental study, which when investigated more fully, should throw light on many problems concerned with the growth of economic plants. Certain physiological and nutritional problems would doubtless be more clearly understood if more were known of the relation between cell division and enlargement and organ or body size. This field of study, in its application to agricultural research, has evidently been little investigated.

Little is now known of the relationship between genetic constitution and cell and tissue activity in any of the economic plants, except in very general terms. Such genetic interrelationships of cells and tissues within a plant organ are of primary concern in any detailed histological study of the growth phenomena in plants.

Recent studies on the relation between cell size and organ size have been concerned principally with fruits. Sinnott (*4*),[3] however, in a study of *Acer* petioles presented results showing that organ size was to some extent directly proportional to cell size in this plant part. He also reviewed several of the earlier investigations on this subject, which show considerable inconsistency especially in general conclusions. Recent morphogenetic studies with fruits, such as those of Houghtaling (*1*), Sinnott (*6*), and Tukey (*7*), suggest the problem of similar growth phenomena in stems and other organs.

The study here reported was made to determine the relation between cell size and number and stem and tissue sizes in two plants closely related genetically but greatly different in stem volume, the cabbage (*Brassica oleracea* var. *capitata L.*) and the kohlrabi (*B. oleracea* var. *caulo-rapa* DC). These plants are both horticultural crops and they are of economic importance largely because of altogether different morphological developments. The terminal bud becomes the economically important or edible part of the cabbage plant; the enlarged stem becomes the edible part of the kohlrabi. The two plants are similar in their seedling stage but can be distinguished by leaf characters beyond that stage.

Based on a genetic study of cabbage and kohlrabi stems, Pease (*3*) concludes that the enlarged stem of the kohlrabi is determined by three multiple factors, two of which are major, the other, a minor or modify-

[1] Received for publication February 19, 1940. The work on which this report is based was done in the Department of Botany, Columbia University, New York.

[2] The writer is grateful to Dr. E. W. Sinnott, of Columbia University, for many helpful suggestions throughout the course of this investigation and to the staff of the Department of Botany, Columbia University, for the use of its greenhouse and laboratory facilities.

[3] Italic numbers in parentheses refer to Literature Cited, p. 470.

ing one. He presents no data, however, on the morphogenetic relationships of these plants, and there seems to be no literature bearing on this specific subject.

MATERIALS AND METHODS

Plants of the White Vienna variety of kohlrabi and the Copenhagen Market cabbage were used in this study. They were grown in the greenhouses of Columbia University during the fall and winter of 1939–40. Throughout most of the growing period it was necessary to lengthen days to 12 to 14 hours by the use of 500-watt lights in order to secure satisfactory thickening of the kohlrabi stem and to prevent excessive etiolation.

It is essential in such studies as these that the data be secured from comparable positions on the plant throughout growth. Under the conditions of this study, the main axes of both plants grew in height at about the same rate and reached about the same height when the rate of growth in height decreased sharply. This decrease in rate of growth indicated the time of beginning of rapid stem enlargement of the kohlrabi and relatively slow stem enlargement but rapid terminal bud development of the cabbage. The first specimens were collected at this stage of development. After considerable preliminary examination, it was found that a position about 20 mm. from the apex was most satisfactory for stem measurements in both plants throughout their development. Thereafter, studies of the growth relationships were made at very nearly that position, which was at about the center of the mature enlarged kohlrabi stem. The data presented were taken from a study of 121 kohlrabi and 48 cabbage plants. The diameter of stem of kohlrabi ranged from 1 mm. to 90 mm., the cabbage stem diameter ranged from 1 mm. to about 12 mm. All the material represented the stages from shortly after differentiation of primary tissues to approximate maturity of the stems. Because of the apparently unavoidable variation in individual plants, it seems necessary in such studies to use a large number of specimens. Uniform environmental factors are also essential.

The plant material was killed, embedded in paraffin, and cut into transverse and longitudinal sections for certain phases of the study. Most of the investigation, however, was carried out by the use of free-hand sections stained with Delafield's haematoxylin. The sections were then temporarily preserved in glycerin for examination and drawing. This latter method proved much more satisfactory, since a large number of specimens could be examined, and also because of the lack of any distortion of cells or cell walls.

The transverse plane was used in determining cell size and tissue size. Since the cells were nearly isodiametric the volume of the cells could be compared with the volume of the tissues or the stem in such a structure as the enlarged kohlrabi. A more accurate method, however, seemed to be to compare the cell diameters with the tissue and stem diameters, since the growth relations in the cabbage and kohlrabi could in this way be compared directly.

Measurements were made of the cell, tissue, and stem diameters by the use of a microprojector. These measurements were made at no less than three levels within 2 to 3 mm. of each other and averaged. Cell measurements were made in the following manner: 20 to 30 cells

in each tissue to be used were drawn under the projector and then 20 of them in each section were measured in two directions. The average of these cell diameters, carefully selected as typical, seemed reliable for this purpose. There was some variation in apparent cell sizes throughout the growth, and those measured represented the average maximum sizes shown.

The development of cell, tissue, and organ size is an exponential process; hence it is more correct to plot the growth values logarithmically. In this study, most of the plotting was done by placing the actual values on double logarithmic paper, but it was found that very wide ranges of values could be most clearly presented by the use of logarithms of actual values.

RESULTS

GENERAL STRUCTURE

The kohlrabi and cabbage stems were evidently similar in anatomy during the early stages of development. The cells within 12 mm. of the terminal meristem were almost entirely responsible for the growth in height of the plant. Those closer than 5 to 7 mm. divided rapidly; those from about 7 to 12 mm. elongated considerably. The region of differentiation seemed to lie between 12 and 16 mm. from the tip. As previously mentioned, the growth concerned in diameter relationships could best be measured at a position about 20 mm. from the tip. At 15 to 20 mm. from the terminal meristem, the pith in both plants made up 70 to 90 percent of the total stem diameter up to a diameter of 4 to 5 mm. The vascular bundles were arranged around the outside of the pith. An active cambium could be readily detected in the bundles, and in most stems there was a less active interfascicular one. The cortex, like the pith, was composed of thin-walled parenchyma during its early development. Later, collenchyma developed in the outer portion, and at near maturity, especially in the kohlrabi, an active periderm was formed in the outermost layers. The anatomy of the kohlrabi stem has been described in some detail by Vöchting (*8*), and it is not necessary to do so here. The writer's observations agreed with Vöchting's in all details noted.

The most striking difference between the structure of the cabbage and kohlrabi stems occured at a stem diameter of 5 to 6 mm. At this stage an inner meristematic region developed in the center of the kohlrabi pith. Shortly after this development, four to six small vascular bundles were laid down in a circle about the group of rapidly dividing central pith cells as seen in transverse sections. As growth continued, these bundles could be seen traversing the pith in all directions, as described by Vöchting (*8*). Rarely were they directly associated with the larger stem bundles already mentioned. Neither this inner meristematic zone nor the medullary bundles were ever found in several other closely related plants examined, including the cabbage, cauliflower, broccoli, and brussels sprouts. In the kohlrabi they first appeared as more or less irregular strands of meristematic cells extending through the central pith. Shortly after their appearance or about the time the ring of medullary bundles was formed, the rapidly dividing cells made up a zone about 1 mm. in diameter as seen in cross section. The zone of cell multiplication enlarged toward the periphery of the pith as growth of the plant continued. At maturity it extended to

within 1 to 2 mm. of the periphery of the pith and thus made up most of the volume of the pith zone. Meristematic activity here seemed always to precede the formation of the pith bundles.

The total pith diameter made up about 90 percent of the kohlrabi and 80 percent of the cabbage stem at maturity. The edible portion of the kohlrabi was, therefore, largely of pith tissue. The bundles and cortex made up the remaining volume in about equal proportions. This study was mainly concerned with the pith, although certain relationships are given for other tissues.

GROWTH RELATIONS IN PITH OF KOHLRABI AND CABBAGE

A very definite relationship could be established between cell size and tissue size at several regions in the pith of the kohlrabi and cabbage stems by careful selection of sections at comparable positions on each plant. It is sometimes desirable to establish volume rather than diameter relationships, but here, because of the decreasing cell size as the apex is approached, it was felt that simple diameter relations at the given position (about 20 mm. from the tip) would be most accurate and comparable. The growth of the cabbage pith seemed to be mostly in width or diameter, since there was little or no increase in length at the position measured, and there was apparently only periclinal and transverse division of cells. Since the largest cells of the pith of the kohlrabi stem were nearly isodiametric and the shape of the entire stem was similar to a sphere, volume relations could be established on that basis if an accurate measure of average cell volume could be made. No method was known, however, by which an accurate volume could be obtained for cells which varied in size in different parts of both the pith and cortex and in shape during division and enlargement. Certain linear relationships, based on diamter at a definite position, could be established, however. The results from a large number of measurements show a somewhat simple growth pattern.

In order to secure a measure of the relative amount of cell division and cell enlargement during the growth of the pith, several types of measurements were made. Some of the relationships are shown in figure 1. The cell diameters of the central pith zone and the peripheral pith region were plotted against the total pith diameters on a double logarithmic grid for both kohlrabi and cabbage. The changes in relative size of the cells and the tissue can then be easily determined by measuring the ratio between the cell size and pith size at numerous stages during increase in size of the pith. This relationship can be expressed by use of the value of k,[4] the constant of relative growth as used by Huxley (*2*). If the change in relative size of cell and pith is constant and the values at different sizes are plotted against each other logarithmically, a straight line will be formed. The slope of this line which can be measured by the value of the constant, k, denotes the relationship between the two size variables. For example, if cell size is increasing at the same rate as organ size, then the line slopes upward at an angle of 45° and the value of k is equal to 1.0. If the cell size increases more slowly than the organ size, k is less than 1.0, and its

[4] From Huxley's formula for measuring the relationship between the magnitudes of two variables. Where x is the value of the organ and y that of the varying part, the relation between them is $\log y = \log b + k \log x$, where b and k are constants. b indicates the value of y when $x = 1$, and is of no importance here. The value of k denotes the relation between the two variables. This simply means that if two variables which follow this formula are plotted against each other on a double logarithmic grid their values will fall in straight lines.

value measures the relation between the two variables. If cell size is not increasing as rapidly as organ size, the difference in the relative value between them must be due to cell division. The relative amount of cell division to cell enlargement can then be calculated from the value of the relative growth constant or the relation between these two variables, or k. These relationships are, of course, independent of time and of absolute rate of growth.

As may be noted (fig. 1, *a*, *b*), growth in the central pith zone in both the kohlrabi and the cabbage was due to both cell division and cell enlargement. Cell division was responsible for most of it, however, up to a total pith diameter of about 2.2 mm., since up to this point the relative growth constant k is equal to 0.2 to 0.3. In other

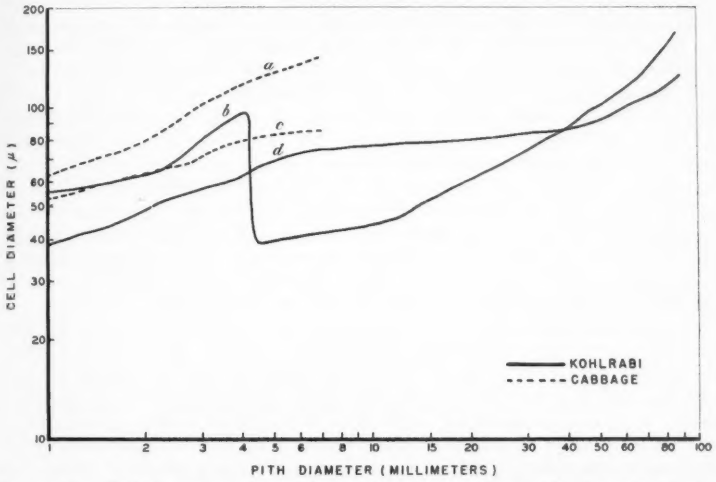

FIGURE 1.—Cell diameter plotted logarithmically against pith diameter during development of the kohlrabi and cabbage stems: *a*, *b*, Central pith cells; *c*, *d*, peripheral pith cells. The sharp break in *b* at a pith diameter of 4 to 5 mm. shows the cell relationships there at the time of development of the inner meristematic zone.

words, 20 to 30 percent of the growth was due to cell enlargement, the remainder, to cell division. At the pith diameter of about 2.5 mm., the kohlrabi central pith showed a rather sharp increase in relative rate of cell enlargement which extended to a total diameter of about 4.2 mm. At the 4.2-mm. diameter, cell enlargement was evidently responsible for about 75 percent of the growth. Cell division rate decreased at about the same stage in the cabbage, but up nearly to maturity it was responsible for about 50 percent of the increase in diameter of the pith (fig. 1). There was no sharp inflection point or position where cell division stopped and cell enlargement began.

The central meristematic zone already described appeared in the kohlrabi pith when it was between 4.0 and 4.5 mm. in diameter. With its appearance there was a sharp decrease in the average cell diameter in this region, as shown in figure 1, *b*. Thereafter, up to a

pith diameter of about 12 mm., cell division was responsible for about 80 percent of the growth in relation to the tissue as a whole. As growth continued, cell division was accompanied by more and more enlargement after each division, until at between 70 and 90 mm. in total diameter, the inner pith cells were enlarging at the same rate as the pith diameter, and $k=1.0$. Cell division evidently had then ceased, and the growth was due entirely to cell enlargement here.

The peripheral pith cells of both the kohlrabi and the cabbage were measured in successive stages of stem development in order to compare their relative rate of cell division and enlargement with that of the central cells just described. The peripheral pith cells measured were those from about the fifth to tenth cell removed from and inside the vascular bundles. The same general relationships appeared in both the kohlrabi and the cabbage up to the time of development of the inner meristematic pith zone in the kohlrabi, or to a pith diameter of about 4.5 mm. (fig. 1, *c*, *d*). Up to this point in the kohlrabi, and throughout the growth of the cabbage, the relative rate of cell division to cell enlargement was greater in the peripheral zone than in the central one. In other words, growth due to proportionately more cell enlargement occurred first in the center of the pith. From the center toward the periphery, there was a definite gradient in this type of growth which could be observed readily by microscopic examination at increasing size of the tissue. The difference in rate of cell division in proportion to cell enlargement in the two zones is well illustrated by the constantly greater value of k (fig. 1) for the central pith of the cabbage and during the early growth of the kohlrabi.

The general relationship just described was upset somewhat by the development of the central meristematic zone in the kohlrabi pith. Evidently, 60 to 70 percent of the growth at the periphery of the kohlrabi pith was due to cell division until a pith diameter of approximately 6 mm. was reached. After that size was attained there was very little change in cell size or in relative rate of cell division until rapid cell enlargement took place soon after the period of rapid enlargement of cells in the central pith just before maturity (fig. 1). The peripheral pith cells then remained larger than the inner ones throughout most of the period of enlargement of the kohlrabi stem. This is evidently a very unusual situation in plant tissues, however. In fact, examination of the cells included in the central meristematic zone showed that within it the largest cells were usually found nearest the center.

RELATIVE NUMBER OF CELLS

The regression line in figure 1 shows the relative changes in cell size at increasing kohlrabi pith size, and from these one may see where increase in cell number must be taking place most rapidly in proportion to pith size, as already described. Nevertheless, in order to clarify the relative changes in cell number, the actual number of cells can be plotted against the pith volume of the enlarged stem or "knob" of the kohlrabi (fig. 2). Because of the extreme range in number of cells, as well as in pith volume, it was more convenient to plot the logarithms of the values rather than the actual values. The measurements were made, their logarithms located on the graph paper, and then regression lines drawn through them as in the other figures.

The number of cells at any stage in the growth of the kohlrabi pith can be determined approximately by dividing the average volume of the cells into the pith volume. To determine the average volume of the pith cells, three regions were used: The center of the pith, the outermost part of the developing meristematic pith region (after it appeared), and the peripheral pith zone. The average cell volume for each was calculated on the basis of a sphere. Then a weighted average cell volume was calculated, based on the respective volumes of the inner and peripheral pith zones. The cell volume thus determined was then divided into the total pith volume, based also on a sphere. The results from 121 plants used in these determinations fit into a linear regression line (fig. 2). The logarithms of the actual values are shown, since cell number and organ volume are more nearly logarithmic or multiple, than additive relationships. Because of the

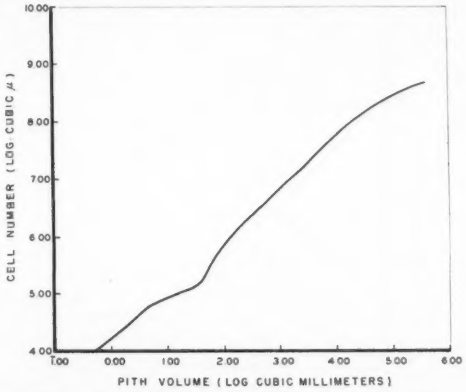

FIGURE 2.—Logarithm of cell number plotted against logarithm of pith volume in the kohlrabi stem during development of the edible portion. The sharp increase in relative cell number at a pith volume of about 1.7 marks the beginning of the rapid cell division in the inner pith.

method of calculation, these results are not as accurate as those presented in figure 1. Nevertheless, the line shows that cell number increases rapidly in the early stages of growth and increases more slowly when the pith is between 2.5 and 4 mm. in diameter. In the later stages, the advent of the central pith cell divisions was indicated by the sharp increase in cell number. The rate of increase was thereafter fairly constant until near maturity, when there was a gradual reduction in rate of increase in number of cells.

The changes in cell number in the cabbage were calculated on the basis of the diameter, that is, the number of cells across the pith in proportion to the diameter of the pith and to that of the stem (fig. 3). The line in this figure which compares the cell number to the diameter of the pith is more accurate, since the pith was growing more rapidly than the stem as a whole. If they had been growing at the same relative rate, either method of comparison would have been satisfactory. As may be noted (fig. 3), the number of cells increased at a fairly constant rate throughout the growth of the stem.

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CELL RELATIONSHIPS IN THE CORTEX

The relationship between the size of the cortical cells and the size either of the organ as a whole or of the tissue itself was so variable that a graphic presentation of the results would not be very reliable. The cells varied in size considerably in different plants of the same diameter and also at different positions in the same plant. Nevertheless, measurements of cell and tissue diameters were made here with as much accuracy as feasible by the same method as used in the pith.

The cortex increased from a radius of 0.2 to 0.3 mm. when the stem was about 4 mm. in diameter to a radius of 1.5 to 2 mm. when the stem of the kohlrabi was 90 to 100 mm. in diameter. The same relationship was shown in the cabbage, but growth of the cabbage stem was limited to 12 to 14 mm. in diameter. There was much less in-

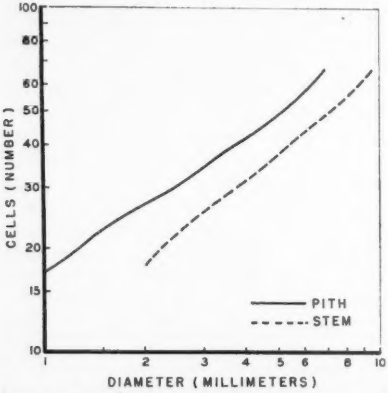

FIGURE 3.—Cell number plotted against pith and stem diameter during the increase in diameter of the cabbage stem. The number of cells represents those in a cross-section or diameter of the pith.

crease in cell size with increase in tissue size than in the pith cells. The average diameter of the cortical cells increased from about 30μ to 40μ during the development of the kohlrabi stem. Periclinal and transverse cell division persisted to maturity. The cabbage plants showed the same relationships in proportion to the diameter of the stem.

RELATIVE GROWTH RATE OF PITH AND ORGANS

The pith diameter was plotted against the total stem diameter at increasing sizes of the kohlrabi and cabbage stems in order to determine the relative growth rates of the pith and stem in these plants (fig. 4). Sinnott (*5*) presented results which showed that in the species he studied, the pith increased more rapidly than the remainder of the stem and that in progressively larger stems "the pith assumes an increasingly greater share of the total size and tissues outside it a smaller and smaller one." Since the pith in kohlrabi included 65 to 70 percent of the diameter of the stem when it was 3 to 4 mm. in diameter, and 90 percent of it at maturity, it is evident that the pith

increased in size more rapidly than the body or stem. Nevertheless, the relative value of this difference in the growth rate of the pith and the stem as a whole was not great. As pointed out by Huxley (*2*), comparatively small variations in the value of k, when plotted on a logarithmic scale, will give large differences if growth continues over a great range in size.

The value of k was determined at several regions along the regression lines (fig. 4) and for the lines as a whole. In all cases, k assumed

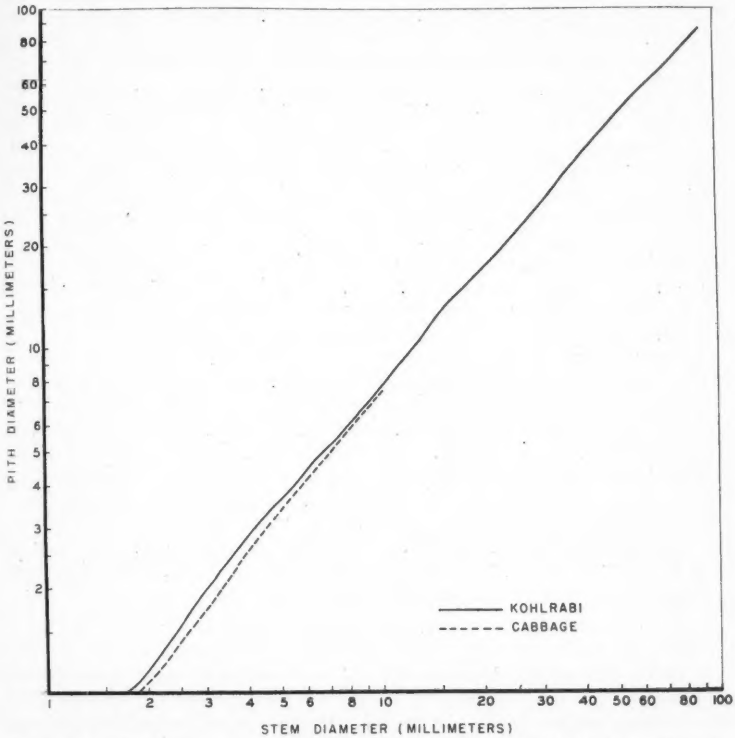

FIGURE 4.—Pith diameter plotted (logarithmically) against total stem diameter of the kohlrabi and cabbage. Both regression lines show k values of very nearly 1.1 throughout the growth of both stems.

a value of very nearly 1.1. This means that the pith was growing 1.1 times as rapidly as the stem as a whole. It might be supposed that the development of the central meristematic pith zone in the kohlrabi would increase its rate of growth in relation to the entire stem, since this zone precedes the formation of the "knob" or edible portion of the kohlrabi, which is composed of about 90 percent pith at maturity. This was not the case, however. It is especially significant that the value of k remained the same here throughout the growth of the stem in spite of the great increase in rate of cell division at the pith

diameter of about 4.5 mm. It is also significant that the growth constant k is very similar for both the cabbage and the kohlrabi (fig. 4); that is, the regression lines slope upward at the same angle. The difference, insofar as these tissue relationships are concerned, was simply in the size of the stem, not in changes in pith and stem growth relationships. In both plants, therefore, the growth of the pith has the same positive and constant differential growth relationship to organ or stem growth. In other words, the cabbage pith might become as large as that of the kohlrabi without change in its growth relationship to the entire stem; it is just a matter of extent of total growth.

GRAFTING TESTS WITH KOHLRABI AND CABBAGE STEMS

Several grafting tests were made to determine whether it was possible to affect the cell relations of either cabbage or kohlrabi stems by interchange and substitution of plant parts. For instance, it was especially desirable to know whether a cabbage stem would assume any of the characteristics of the kohlrabi if the upper portion of a plant, or that bearing the leaves, was of kohlrabi and the lower portion of the stem was of cabbage, and also whether there would be any transmission of cell and tissue characteristics if an approach graft of the stems of the two plants was made. In this way the cells and tissues of one plant could be caused to grow for a considerable distance along the stems in direct contact with those of the other plant.

It was found that cabbage and kohlrabi stems could be grafted rather easily. Several types of grafts were made at different stages of growth and at different positions on the plants. It is only necessary to state here that in no case did either plant seem to affect the cell or tissue relations in the other. In other words, the cells of each plant retained their own morphogenetic nature regardless of their relationship to cells of another genetic character. These results are, perhaps, not surprising to those who are familiar with stock and scion relations in horticultural material. Nevertheless, they seem worthy of note in view of the recent work on the relation of transfer of certain growth substances to cell activity in plants.

DISCUSSION

The results of studies of kohlrabi and cabbage stem growth are significant because the plants, which are closely related genetically, were found to be similar in certain tissue growth relationships but markedly different in others. The decrease in relative rate of cell division, first in the innermost part of the pith and then gradually toward the periphery, is notable, since the same general trend has been noted in petioles (*4*) and cucurbit fruits (*6*). As in these fruits, there was much increase in cell volume, especially in the kohlrabi pith, even during the period of cell division. The inflection point marking the end of cell division and the period of cell enlargement was not so clearly marked in kohlrabi and cabbage stems, however, as was noted in cucurbit fruits. In general, however, there was a gradual decrease in the relative rate of cell division to cell enlargement as the stems increased in size. Thus these observations and calculations show that in this pith tissue there was more and more cell enlargement after each cell division.

The development of the central meristematic zone in the pith of the kohlrabi seems remarkable, since this zone is the first to show a decreasing rate of cell division in the early stages. The medullary bundles are evidently not concerned with the initiation of this zone, since rapid cell division begins before these bundles are laid down. It is significant that as soon as these innermost cells reach a diameter of about 95μ they begin rapid division. During the growth in size of the pith, the cells farther and farther removed from the center reach that diameter, and then they start rapid division. The most peripheral pith cells do not reach 95μ until very late in the development of the stem, and this slow increase in size may be the reason why these peripheral pith cells never become extremely meristematic, as do the more central cells. The basic reason for the early difference in relative rates of cell division to cell enlargement between the central and peripheral pith cells is not clear.

In the cabbage, rate of cell division decreases first in the central pith, then in successively more peripheral cells, as in the early stages of kohlrabi growth. Its activity persists longer in the extreme peripheral pith and in the cortex (considering only the primary tissues). These results suggest that perhaps some growth substance or physiological factor associated with the bundles might be responsible for the delay in maturity of nearby cells. Such a factor would not explain the condition in the pith of the older kohlrabi, however, where the central meristematic zone develops.

That there is no relationship between cell division and total growth of pith size in proportion to stem size is well illustrated in this study. In spite of the advent of the very meristematic zone in the kohlrabi pith, the same rate of growth relative to the stem as a whole obtains here as in its earlier growth, and as in the cabbage stem. The morphogenetic difference between the kohlrabi and the cabbage stems seems to be due largely to the persistence of cell division, accompanied by cell enlargement, to a greater stem size before growth ceases. The difference is also partly due to the greater size finally attained by the kohlrabi pith cells.

It may be concluded that there are several morphogenetic factors involved in the development of size differences in the stems of closely related plants. Thus, the genetic factors concerned with histological development are responsible for several definite structural differences between the cabbage and kohlrabi stems. Some of these can be measured by a quantitative analysis of structural changes during the course of development.

It would seem that a more fundamental solution to many horticultural problems could be obtained if more were known of the histological development of the plants or plant parts concerned. More might be known also about the influence of different practices or treatments on the relative rates of cell division and cell enlargement and on tissue and organ size relationships.

SUMMARY

In developing kohlrabi and cabbage stems cell diameter in the pith and cortex was compared with pith and also with stem diameters.

The greater size of the kohlrabi stem was due largely to a greater number of cells, although some of the difference was evidently due to cell size.

The pith, of which the edible kohlrabi is largely composed, grew at the same rate, relative to the entire stem, throughout its development, and at the same relative rate as that of the cabbage pith to its stem.

There was no sharp point of demarcation between the cell division stage and the cell enlargement stage in the growth of the pith and cortex of these stems.

The innermost cells of the pith increased most rapidly in size, and, in the kohlrabi, were the first to become very meristematic again after a definite size was reached.

By use of grafting tests with kohlrabi and cabbage stems, it was found that the cells of each plant retained their own morphogenetic nature even though they developed in direct contact with each other.

Some factors involved in the histological differences between the stems of the two plants are discussed.

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THE APPARENT DIGESTIBILITY AND NUTRITIVE VALUE OF BEARDLESS WHEATGRASS AT THREE STAGES OF MATURITY [1]

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INTRODUCTION

The digestibility and nutritive value of range grasses in the green, immature stages have not been studied extensively by research workers and there is little information available concerning the relative feeding value of range forage at various stages of maturity.

Some studies have been made concerning the changes in the chemical composition of range plants as maturity advances. However, the chemical composition is not a true index of the nutritive value, partly because of differences in coefficients of apparent digestibility of the various organic nutrients and because of certain other differences such as those relating to palatability.

The study reported in this paper was made to determine the chemical composition and the coefficients of apparent digestibility of the organic nutrients of beardless wheatgrass (*Agropyron inerme* (Scribn. and Smith) Rydb.) at a 3- to 5-inch stage, a 7- to 10-inch stage, and a headed stage of growth. These three stages of development will hereafter be referred to as the early, medium, and late stages, respectively. Beardless wheatgrass is an important native range bunchgrass in the State of Washington and the Pacific Northwest. It is quite similar to bluebunch wheatgrass (*A. spicatum* (Pursh) Scribn. and Smith) except that the former, as the name implies, is awnless, whereas bluebunch wheatgrass is awned or bearded.

REVIEW OF LITERATURE

SEASONAL VARIATION IN CHEMICAL COMPOSITION

Much of the work concerning the seasonal variation in chemical composition has been done with tame grasses. This review includes papers which deal only with range grasses.

McCall (*11*),[3] in studying the chemical changes of bluebunch fescue (*Festuca idahoensis* Elmer) clipped at semimonthly intervals, found the protein content declined from 25.55 percent in the young growing grass to 4.56 percent in the mature grass 4 months later. The amount of crude fiber increased as the plant matured and nitrogen-free extract reached its highest point at maturity. The values for crude fat showed a tendency to decline as maturity advanced and crude ash values showed the opposite trend. The highest percentages of calcium and phosphorus were found in the young grass. The phosphorus

[1] Received for publication February 26, 1940. Published as Scientific Paper No. 449, College of Agriculture and Agricultural Experiment Station, State College of Washington.

[2] The author wishes to express his appreciation for helpful suggestions and criticisms to Jerry Sotola and H. Hackedorn, of the Washington Agricultural Experiment Station, and to E. H. Steffen, of the college faculty.

[3] Italic numbers in parentheses refer to Literature Cited, p. 478.

content declined more rapidly than the calcium content as maturity advanced.

Seasonal changes in the chemical composition of blue grama (*Bouteloua gracilis*), hairy grama (*Bouteloua hirsua*), and curly mesquite (*Hilaria belangeri*) were studied by Stanley and Hodgson (*17*). Amounts of moisture, crude protein, and phosphorus were high in the young plants and decreased as the plants matured. The percentages of total ash and the ether extract content showed no definite trends. Amounts of nitrogen-free extract and crude fiber increased as maturity advanced. The percentage of calcium was highest when the plants were young.

Gordon and Sampson (*7*), working with graminaceous, grasslike, and broadleaved herbaceous species of foothill range plants, found a rather orderly decline in the percentage of crude protein, a silica-free ash, calcium, phosphorus, and potassium from early growth to plant maturity. The percentage of crude fiber increased as maturity advanced. The most rapid changes in composition occurred from the early leaf stage to the full-bloom period.

Hart, Guilbert, and Goss (*8*) found that the dry matter of California range forages varies from that of a protein-rich concentrate during the early vegetative stages to that of a poor roughage during the drought period.

The carotene content of black grama (*Bouteloua eriopoda* (Torr.) Torr.) and mesa dropseed grass (*Sporobolus flexuosus* (Thurb.) Rydb.) was found by Watkins (*18*) to be highest during the early growing season.

Smith and Stanley (*14*) tested blue grama grass (*Bouteloua gracilis*) at three stages for vitamin A by the rat-growth method, and found that the young grass was a very potent source of vitamin A. There was only about one-half as much vitamin A in the grass at maturity, and after weathering had taken place the amount was negligible.

According to Atkeson and his coworkers (*4*) little bluestem (*Andropogon scoparius*), big bluestem, (*Andropogon furcatus*), and buffalo grass (*Buchloë dactyloides*) have their highest carotene content in June. When growth ceases the carotene content quickly decreases.

DIGESTION STUDIES

Kennedy and Dinsmore (*9*) conducted one of the early digestion experiments with green range forages. They found the crude protein to be 68.0 and 63.9 percent digestible for western bromegrass (*Bromus marginatus*) and native bluegrass (*Poa sandbergii*) with nutritive ratios of 1:8.5, and 1:8.7, respectively. The grasses were headed when fed.

In an early experiment with a Guernsey cow fed mixed pasture grass, Armsby and Caldwell (*2*) found that the digestibility of all the nutrients decreased materially as the forage matured.

Woodman, Blunt, and Stewart (*21*, *22*) state that early grass is rich in digestible protein and is, in effect, a "watered" protein concentrate. They found that crude fiber in young pasture grass was about 80 percent digestible. The organic matter of pasture grass was about 10 percent less digestible when grazed in June than when grazed a month earlier, according to Woodman and coworkers (*23*).

The dry matter in fresh young grass was 74.4 percent and crude fiber 80.4 percent digestible as determined by tests with two sheep by

Watson and Ferguson (*20*). Newlander and Jones (*13*) found the dry matter, protein, fiber, nitrogen-free extract, and ether extract of fresh young grass to be 71.0, 76.5, 74.4, 77.9, and 40.0 percent digestible as determined with dairy cows.

Christensen and Hopper (*5*) found that the digestibility of mature prairie hay consisting of 50 to 75 percent of western needlegrass (*Stipa comata*) was considerably higher when cut in July than when left until October.

McCall [4] studied the digestibility of mature bluebunch wheatgrass (*Agropyron spicatum*) which was gathered in October The coefficients of digestibility as determined with six sheep were 36.0 percent for the dry matter, 50.6 percent for the crude fiber, 38.8 percent for nitrogen-free exact, and 32.0 percent for crude fat. The digestion coefficients for crude protein were negative and there was an average negative nitrogen balance of 17 gm. per sheep for the 10-day period. On an 88.76-percent dry-matter basis, the bluebunch wheatgrass contained 34.23 percent total digestible nutrients.

Crampton and Forshaw (*6*), working with rabbits, studied the intraseasonal changes in the nutritive value of pasture herbage clippings consisting of Kentucky bluegrass, redtop, and wild white clover, cut at 10-day intervals, and found "a progressive decline in growth-promoting value and digestibility of herbage from spring until midsummer and a complete recovery in both respects in the fall-growth material." There was a gradual decrease in digestibility of dry matter, nitrogen, cellulose, and nitrogen-free extract as the season advanced, followed by a complete recovery in the fall months.

EXPERIMENTAL METHODS AND PROCEDURE

Crossbred range yearling sheep from Lincoln × Merino ewes and Hampshire rams were used. The individual average weights ranged from 38 to 53 kg. (84 to 117 pounds). Each feeding period consisted of 10 days and was preceded by a 3-day preliminary period. A description of the metabolism cages and methods employed has been given by Sotola (*15*).

Grass was clipped daily with sheep shears from native stands near Colton, Wash., of almost pure beardless wheatgrass which had never been plowed or cultivated. The bunches were clipped as close to the ground as was possible without injuring the crown of the plants. The grass was fed green and the sheep given all they would consume. Five grams of salt containing 0.02 percent of potassium iodide was fed daily, and water was available at all times.

The trials on the medium and late grasses were conducted in the spring of 1938 and the early stage in the spring of 1939. The trial of the late stage was begun as soon as the grass was well headed and before it started to become dry. The anther-falling stage occurred when the late digestion trial was about half over. Analytical procedures as recommended by the Association of Official Agricultural Chemists (*3*) were followed in making the analyses.

[4] McCALL, RALPH. SEASONAL VARIATION IN THE CHEMICAL COMPOSITION AND DIGESTIBILITY OF CERTAIN SPECIES OF RANGE GRASSES. 137 pp., illus. 1932. [Thesis, Wash. State Col.]

CHEMICAL COMPOSITION IN RELATION TO MATURITY

The chemical composition of beardless wheatgrass is given in table 1 on the fresh basis and on the water-free basis. The dry matter of the grass was about 14 percent greater at the late stage than at either of the younger stages. The dry-matter content at the early stage was slightly higher than at the medium stage. This was probably because it was taken from an area which was drier and more rocky than the area from which the grass at the medium stage was clipped. As has been stated, the trials on the two youngest stages were conducted in different seasons, but a study of the rainfall and temperature of the two seasons does not show any appreciable differences in these two factors.

The crude protein percentage decreased materially as the grass matured. There was a slight variation in the crude fat content but the changes were not marked. There was nearly twice as much crude fiber in the grass at the late stage as at either the early or medium stages. This is probably a major factor in the reduction of digestibility as the grass matured. The nitrogen-free extract increased about 5 percent in the late stage, and the difference was slight between the two younger stages. The crude-ash content remained about the same in the younger stages and increased slightly in the late stage on the fresh basis.

TABLE 1.—*Chemical composition of beardless wheatgrass at three stages of maturity*

Basis and state of maturity	Dry matter	Crude protein (N × 6.25)	Crude fat	Crude fiber	N-free extract	Crude ash	Calcium	Phosphorus	P:Ca ratio (P as 1)
	Percent	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	
Fresh grass:									
Early	32.49	6.75	1.32	8.04	13.05	3.33	0.14	0.12	1.17
Medium	31.80	5.25	1.30	8.98	12.97	3.30	.11	.09	1.22
Late	45.51	4.07	1.57	15.51	20.80	3.56	.14	.11	1.27
Dry matter:									
Early		20.79	4.06	24.75	40.16	10.24	.44	.37	1.19
Medium		16.52	4.08	28.24	40.78	10.38	.35	.29	1.21
Late		8.95	3.45	34.07	45.71	7.82	.30	.24	1.25

The highest content of calcium and phosphorus in the beardless wheatgrass coincided with the youngest stage and decreased as the grass matured, with a slight widening of the calcium-to-phosphorus ratio. The content of calcium and phosphorus compares favorably with that found by McCall (*11*) for bluebunch fescue at corresponding stages of maturity. The calcium-phosphorus ratio does not equal the approximately 2:1 ratio found in the bones of animals, but lies within the normal limits of the 2:1 and 1:2 calcium-phosphorus ratios given by Maynard (*10*).

DIGESTION STUDIES

COEFFICIENTS OF APPARENT DIGESTIBILITY

Table 2 contains the coefficients of apparent digestibility as determined with individual sheep, as well as the average for each trial.

The coefficients of apparent digestibility of all the organic nutrients decreased as the grass matured with the exception of the crude fat

which was slightly more digestible at the medium stage than at the early stage. The decline indigestibility was greater between the medium and late stages than between the early and medium stages of maturity. The nutrients at the two younger stages resemble those of a concentrate rather than a roughage. The fat and crude protein decreased most in digestibility as maturity advanced, followed in order by the fiber and nitrogen-free extract.

TABLE 2.—*Coefficients of apparent digestibility of beardless wheatgrass at three stages of maturity*

Stage of maturity	Sheep No.	Sex	Dry matter	Crude protein (N × 6.25)	Crude fat	Crude fiber	N-free extract
			Percent	*Percent*	*Percent*	*Percent*	*Percent*
Early	1	M	72.0	80.0	69.0	77.0	77.0
	3	M	72.0	80.0	61.0	79.0	77.0
	4	M	71.0	78.0	63.0	78.0	74.0
Average			71.7	79.3	64.3	78.0	76.0
Medium	1	F	68.0	75.0	67.0	74.0	74.0
	2	F	68.0	76.0	68.0	76.0	74.0
	3	F	69.0	77.0	67.0	77.0	74.0
	4	F	69.0	76.0	66.0	75.0	75.0
	5	M	67.0	75.0	63.0	73.0	73.0
	6	F	67.0	77.0	65.0	72.0	73.0
Average			68.0	76.0	66.0	74.5	73.8
Late	1	F	62.0	64.0	46.0	66.0	67.0
	2	F	64.0	62.0	46.0	70.0	69.0
	3	F	64.0	66.0	48.0	69.0	68.0
	4	F	63.0	65.0	53.0	66.0	68.0
Average			63.2	64.2	48.2	67.8	68.0

DIGESTIBLE NUTRIENTS

The digestible nutrients and nutritive ratios of fresh beardless wheatgrass at the three stages of maturity are shown in table 3. Although the digestion coefficients of the various nutrients were lower in the late stage than in the immature stages (table 2), the percentage of total digestible nutrients of an equal weight of fresh green grass was about 6 percent larger than in either of the other stages as a result of the larger percentage of dry matter in the late stage.

The difference in total digestible nutrients between the two younger stages was not marked, although the early stage was slightly higher in digestible nutrients than the medium stage, owing to the slightly higher digestion coefficients and the greater dry-matter content. The digestible nutrients depend on the dry-matter content as well as the digestion coefficients and would vary according to the dry matter when expressed on a basis of fresh grass. When computed on a dry-matter basis, the total digestible nutrients were 72.10, 69.85, and 63.62 percent for the early, medium, and late stages, respectively, showing that the digestible nutrients in the dry matter decreased as the grass matured.

There was a marked difference in the quality of the feed at the three stages of maturity as evidenced by the protein content and nutritive ratio. The digestible protein decreased from 5.34 percent in the early stage to 2.61 percent in the late stage. The nutritive ratio increased from 1:3.39 to 1:10.10 in these same stages. When

put on a 10-percent moisture basis, the percentages of total digestible nutrients of the early stage and the medium stage were 7 and 5 percent higher, respectively, than the average total digestible nutrients found by Sotola (*16*) for the leaves of first, second, and third cuttings of alfalfa hay. The nutritive ratios of the grass were slightly wider than for the alfalfa leaves. The total digestible nutrients for the grass at the late stage was about the same as for alfalfa leaves, but the nutritive ratio was about 3.5 times as wide.

TABLE 3.—*Digestible nutrients in fresh beardless wheatgrass at three stages of maturity*

Stage of maturity	Total dry matter	Dry matter	Crude protein (N × 6.25)	Crude fat	Crude fiber	N-free extract	Total digestible nutrients	Nutritive ratio, 1 to —
	Percent	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	*Percent*	
Early	32.49	23.41	5.34	0.85	6.27	9.91	23.43	3.39
Medium	31.80	21.70	3.99	.86	6.71	9.59	22.23	4.57
Late	45.51	28.76	2.61	.76	10.50	14.14	28.96	10.10

CONSUMPTION OF FRESH GRASS AND WEIGHTS AND GAINS OF SHEEP

The daily consumption of fresh grass and the average gains made by sheep during the 10-day digestion trials are presented in table 4. The gains made on the grass at the three stages of growth decreased slightly as the grass matured. However, too much emphasis cannot be placed on the weight records and gains because of the short period involved and the weights being based on 1-day weighings. The sheep also had free access to water before being weighed.

The amount of fresh grass consumed daily decreased with advanced maturity. This decrease was quite marked between the medium and late stages.

TABLE 4.—*Daily consumption of fresh grass, average weights, and gains by sheep fed beardless wheatgrass at three stages of maturity*

Stage of maturity	Average weight of sheep	Average gain by sheep in 10 days	Average daily consumption of fresh grass
	Kilograms	*Kilograms*	*Kilograms*
Early	42.07	1.89	3.10
Medium	43.15	1.78	2.98
Late	46.61	1.36	2.13

CONSUMPTION OF DRY MATTER, DIGESTIBLE NUTRIENTS, AND CALCIUM AND PHOSPHORUS

The average daily consumption of dry matter, digestible nutrients, and calcium and phosphorus per 45.36 kg. (100 pounds) body weight are given in table 5.

The consumption of dry matter decreased materially as the grass matured even though the more immature fresh grass had a much higher percentage of water, necessitating the handling of more bulk by the sheep. Since the percentage of total digestible nutrients of the dry matter decreased as maturity advanced, it follows that the consump-

tion of digestible nutrients was less at the advanced stages of maturity. The differences in the dry-matter intake and different planes of nutrition at the three stages of maturity should have no effect on the coefficients of apparent digestibility, as has been shown by Armsby (*1*), Watson and coworkers (*19*), and Woodman and coworkers (*23*).

The larger consumption of fresh grass and dry matter at the immature stages indicates that the young grass is more succulent and palatable to sheep than grass that is more mature. This should result in a larger consumption of digestible nutrients of a higher quality by animals when grazing on the more immature forage than when grazing on mature forage, with a resulting faster rate of increase in weight and growth. That such is the case is shown by the consumption and gain figures in tables 4 and 5.

TABLE 5.—*Average daily consumption of dry matter, digestible nutrients, and calcium and phosphorus per 45.36 kg. (100 pounds) body weight by sheep fed beardless wheatgrass at three stages of maturity*

Stage of maturity	Dry matter consumed daily	Total digestible nutrients in dry matter	Total digestible nutrients consumed daily	Calcium consumed daily	Phosphorus consumed daily
	Grams	*Percent*	*Grams*	*Grams*	*Grams*
Early	1,072	72.10	773	4.72	3.97
Medium	931	69.85	650	3.26	2.70
Late	865	63.62	550	2.60	2.08

The calcium and phosphorus figures show a rather large daily intake of both of these minerals. The consumption is from two to three times greater than the calcium and phosphorus requirements estimated by Mitchell and McClure (*12*) for growing Shropshire sheep.

NITROGEN BALANCE STUDIES

The data of the nitrogen balance studies during the 10-day feeding period are presented in table 6. A positive nitrogen balance was shown for all three trials. The nitrogen retention was greater for the two younger stages of maturity than for the late stage. The variation for nitrogen intake stored for the individual sheep in the last two trials was quite large, being from 10 to 25 percent in the medium stage, and from 3 to 18 percent in the late stage.

TABLE 6.—*Average results of nitrogen metabolism studies with sheep fed beardless wheatgrass at three stages of maturity*

State of maturity	Nitrogen consumed	Nitrogen voided			Nitrogen balance	Intake stored
		Feces	Urine	Total		
	Grams	*Grams*	*Grams*	*Grams*	*Grams*	*Percent*
Early	331	69	217	298	45	13.60
Medium	241	58	140	198	43	17.84
Late	128	46	70	116	12	9.38

SUMMARY AND CONCLUSIONS

A summary of the results of 10-day digestion trials with yearling sheep fed beardless wheatgrass (*Agropyron inerme* (Scribn. and Smith) Rydb.) at an early (3- to 5-inch) stage, a medium (7- to 10-inch) stage, and a late (headed) stage is presented.

The crude protein content decreased as the plant matured while the crude fiber and nitrogen-free extract increased. The crude fat and crude ash showed slight increases.

The coefficients of apparent digestibility of various nutrients decreased as the grass matured. All the nutrients were highly digestible in the youngest stages. The crude protein was 79 percent digestible in the early stage as compared to 64 percent in the late stage. The decrease in digestibility was greater between the medium and the late stages than between the early and the medium stages.

The dry matter of the grass at the early stage contained 72.10 percent total digestible nutrients while that at the medium stage contained 62.85 percent, and when cut at the late stage 63.62 percent. The nutritive ratios for the three stages were 1:3.39, 1:4.57, and 1:10.10, respectively.

The daily consumption of fresh grass, dry matter, and total digestible nutrients decreased with advanced maturity of the grass. The sheep made larger gains during the digestion trial when fed the immature grass than when fed the more mature forage.

The results of this study indicate that sheep grazing on the range may be expected to consume more total digestible nutrients daily of a higher quality from grass in the early and medium stages than from mature grass, and this should result in a faster rate of increase in growth and weight. A larger area of young grass than of mature grass, however, would be required to produce an equal amount of digestible nutrients.

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U. S. GOVERNMENT PRINTING OFFICE: 1940